Blood Results
in Clinical Practice

A practical guide to interpreting blood test results

For the full range of M&K Publishing books please visit our website:
ww.mkupdate.co.uk

Blood Results
in Clinical Practice

A practical guide to interpreting blood test results

Graham Basten

2nd edition

Blood Results in Clinical Practice
Dr Graham Basten

ISBN: 978-1-910451-16-8

First published 2013
Reprinted 2014, 2017
This revised and updated edition published 2019

All rights reserved. No part of this publication may be reproduced, stored in a retrieval system, or transmitted in any form or by any means, electronic, mechanical, photocopying, recording or otherwise, without either the prior permission of the publishers or a licence permitting restricted copying in the United Kingdom issued by the Copyright Licensing Agency, 90 Tottenham Court Road, London, W1T 4LP. Permissions may be sought directly from M&K Publishing, phone: 01768 773030, fax: 01768 781099 or email: publishing@mkupdate.co.uk

Any person who does any unauthorised act in relation to this publication may be liable to criminal prosecution and civil claims for damages.

British Library Cataloguing in Publication Data

A catalogue record for this book is available from the British Library

Notice

Clinical practice and medical knowledge constantly evolve. Standard safety precautions must be followed, but, as knowledge is broadened by research, changes in practice, treatment and drug therapy may become necessary or appropriate. Readers must check the most current product information provided by the manufacturer of each drug to be administered and verify the dosages and correct administration, as well as contraindications. It is the responsibility of the practitioner, utilising the experience and knowledge of the patient, to determine dosages and the best treatment for each individual patient. Any brands mentioned in this book are as examples only and are not endorsed by the publisher. Neither the publisher nor the authors assume any liability for any injury and/or damage to persons or property arising from this publication.

To contact M&K Publishing write to:

M&K Update Ltd · The Old Bakery · St. John's Street

Keswick · Cumbria CA12 5AS

Tel: 01768 773030 · Fax: 01768 781099

publishing@mkupdate.co.uk

www.mkupdate.co.uk

Designed and typeset by Mary Blood

Printed in Scotland by Bell & Bain, Glasgow

Contents

Preface vii

Abbreviations in the text ix

1 Blood results made easy 1

2 What are we actually measuring and why does this matter? 21

3 Family groups 29

4 Case study: Interpreting abnormal results 35

5 The full blood count 41

6 Coagulation and deep vein thrombosis 49

7 Inflammatory markers 53

8 The immune system 59

9 Transfusion testing 65

10 Chronic disease markers: Diabetes 67

11 Chronic disease markers: Cholesterol 71

12 Chronic disease markers: Chronic obstructive pulmonary disease and acid base 75

13 Thyroid function 79

14 Bone profile 83

15 Renal function: Urea and electrolytes 87

16 Liver function tests 95

Afterword 101

Further reading and references 103

Index 105

Preface

The second edition of this book continues to use storytelling to aid understanding, and also introduces a new and unique 10-point system to help explain blood results. The use of storytelling has been significantly improved and refined following several years of feedback on the first edition.

The purpose of the book has also evolved. Given that so many protocols and decisions can now be found online, and many NHS trusts and private providers produce similar flow charts, the character of the book has been adjusted: it is less formal than a biochemistry textbook while still containing more narrative than an online protocol. It provides an excellent, accessible introduction to blood tests and what they mean. It also enables advanced practitioners to reflect on and improve their practice, and includes new and updated sections of relevance to physiotherapists, paramedics, pharmacists and advanced nurse practitioners.

The main audiences for the book are:

1) Undergraduate or postgraduate healthcare students

2) Healthcare professionals who need an essential handbook for quick reference or as an accessible text for a new area of practice (for example, new MSK First Contact or paramedic practitioners in primary and community care)

3) Patients, clients, friends and relatives who may wish to know more about what a particular blood test means.

Throughout the book, storytelling is used. For instance, we will talk about 'fire engines' and 'police cars' to help explain what are essentially abstract concepts. When a storytelling device is being used, this will be flagged up in the text as '**storytelling**'.

The human body's organs and functions are incredibly complicated and interlinked. The book will therefore outline such links in terms of '**family groups**'. For example, the kidney is responsible for supporting the production of red blood cells; when looking at the full blood count (FBC), it may therefore be helpful to look at the renal or kidney function. These connections are highlighted in the text by the LINK ◀▶ symbol.

Blood tests seldom provide a diagnosis on their own; they are best used in conjunction with case history, X-rays, scans and other reports by allied health professionals.

Given that reference ranges are specific to the machine which analyses the blood in a particular healthcare setting, no formal ranges are presented in this book. You must only use the range presented alongside the result or the range approved by your local healthcare setting.

The values and interpretations used in the book are based on current national guidance. Readers should always seek definitive local approved guidance on diagnosis, treatment, additional blood tests to request and file, and repeat timeframes. Additional resources can be found in the Further Reading section.

The main aim of the book is to demystify what some consider to be a complex and difficult topic. I've been teaching this stuff for over 10 years to a wide range of students and delegates within UK universities, the NHS and private providers. I get great pleasure from knowing that, in some small way, this book has helped improve the patient's or client's journey.

Have fun, enjoy it and let me know how it's helped.

Graham P Basten PhD MIBMS FHEA

Abbreviations in the text

ACTH	adrenocorticotropic hormone
AFP	alpha-fetoprotein
AlkPhos	alkaline phosphatase
ALT	alanine aminotransferase
ANCA	anti-neutrophil cytoplasmic antibodies
aPTT	activated partial thromboplastin time
AS	ankylosing spondylitis
AST	aspartate aminotransferase
BNP	B-type natriuretic peptide
BPH	benign prostatic hyperplasia
CEA	carcinoembryonic antigen
CDL	clinical decision limits
CHD	coronary heart disease
CKmb	creatine kinase mb
COPD	chronic obstructive pulmonary disease
COX-2	cyclo-oxygenase 2
CRP	C-reactive protein
CSWS	cerebral salt-wasting syndrome
CVD	cardiovascular disease
DVT	deep vein thrombosis
EDTA	ethylene-diamine-tetra-acetic acid
eGFR	estimated glomerular filtration rate
ESR	erythrocyte sedimentation rate
FBC	full blood count
FN	false negative
FP	false positive
GGT	gamma-glutamyl transferase

Hb	haemoglobin
HbA1C	haemoglobin with glucose irreversibly bound
Hct	haematocrit
HDL	high-density lipoprotein
HepBsAg	Hepatitis B surface antigen
HIV	human immunodeficiency virus
INR	international normalised ratio
K	potassium
LDH	lactate dehydrogenase
LDL	low-density lipoprotein
LFT	liver function tests
MCV	mean cell volume
MI	myocardial infarction
Na	sodium
PA	psoriatic arthritis
PE	pulmonary embolism
PMR	polymyalgia rheumatica
PSA	prostate specific antigen
PST	plasma separator tube
PT	prothrombin
PTG	parathyroid gland
PTH	parathyroid hormone
PV	plasma viscosity
RA	rheumatoid arthritis
RBC	red blood cell count
Rh	Rhesus
SIADH	syndrome of inappropriate anti-diuretic hormone
SLE	systemic lupus erythematosus
SPEP	serum protein electrophoresis

SST	serum separator tube
T3	triiodothyronine
T4	thyroxine
TIBC	total iron binding capacity
TN	true negative
TP	true positive
TRH	thyrotropin-releasing hormone
TSH	thyroid-stimulating hormone
U&Es	urea and electrolytes
ULN	upper limit of normal
VTE	venous thromboembolism
VWF	von Willebrand's factor
WBC	white blood cell count
WS	Well's score

1

Blood results made easy

This book will enable you to:

- Understand how blood is split into chemicals (biochemistry) and cells (haematology)
- Look for patterns using family groups
- Consider how blood tests can change cellular content, waste products, production and interaction
- Be more confident when identifying and managing 'out of range' and 'in range' blood tests in a symptomatic patient
- Consider four questions when interpreting the blood result:
  - How far out of range is it?
  - Do we have a clinical decision limit or protocol?
  - Does the result make sense?
  - Does the family group support the result?
- Determine the importance of what is being measured, understand why and when it was requested, and know what to do next with the result?

'Storytelling'

I'd like you to imagine that you are not a healthcare professional, student, client or patient. Instead, imagine that you are a famous detective. (Any will do – you decide.) You have arrived at a crime scene and you will look at various pieces of evidence (symptoms) and will hear several witness statements. Each witness statement represents a blood test result. With each statement, you need to ask:

- How reliable is the witness?
- When was the statement taken?
- Does it make sense?
- How close was the witness to the crime scene?
- Most importantly do their 'mates', friends or associates back up the story?
  (If we get corroborated statements all saying the same thing, we can usually place greater confidence in them.)

We will return to this approach throughout the book.

Key themes

In this section we will explore key themes to help interpret blood test results. As we interpret the blood test results, we can consider some initial questions.

What are we measuring?

If we know what the blood test is actually measuring, that should help us understand the 'so what' question. Is it a cell or a chemical? We can split blood tests into two types – biochemistry and haematology. The former measure all the chemicals in the blood, while the latter measure all the cells in the blood. The biochemistry tests measure liver function, kidney function, inflammation, thyroid, autoimmune and are generally more closely associated with urgent 'red flag' conditions like hyperkalaemia – raised potassium (K). The haematology tests measure cells and report on the types of cells, how many there are and how big they are. This is very useful when we are looking at patterns of anaemia and infection, and 'red flag' conditions like myeloma and leukaemia. (This is all covered in more detail in Chapter 5.)

To learn more about how to separate the blood into these two components, see the section on 'Blood collection' (p. 5) and check local phlebotomy protocols.

Why did we measure it?

Some thoughts under this theme:

- Was the patient symptomatic and did we specifically request the blood test to confirm our thinking?
- Is the result incidental? For example, if the patient is asymptomatic, did they have a routine health screen or pre-operative assessment which highlighted an out-of-range result?
- Are we requesting the blood test as part of a triage exclusion service?
- Is this part of normal pathology management such as drug or disease monitoring?

When did we measure it?

Some thoughts under this theme:

- Did we measure this blood test on a hospital ward 10 minutes ago? Or was it measured in primary care three months ago? (There may be good reasons for both these scenarios.)
- Have the patient's results always been raised or decreased? Despite being out of the normal range, is your patient 'normal' for their cohort?

What do we do next with the result?

Think about the following:

- Do we have a protocol or clinical decision flow chart for results which are out of range?

- What is my remit – to treat or to refer?
- When do I file as 'normal'?
- When do I escalate or de-escalate?

We can summarise these as four key questions.

Question 1 – How far out of range is it?

Consider:

- What is normal in your setting?
- What's the biggest or smallest result you've seen in your setting?
- Do you have clinical limits (see later chapters)?
- Is it always slightly out of range?
- Is it within a range or group that could contain false positive results (see later chapters)?

Question 2 – Does the result make sense?

Consider:

- Has the patient just had an operation?
- Has the patient been started on medication?
- Do they have symptoms?
- In short, do the results match the person in front of you?

Question 3 – What do the family groups tell us? Do they all agree?

See Chapter 3 for more detail on family groups.

Question 4 – Is this an important blood test?

Consider:

- Which are the 'go to' blood tests in your setting, the ones that people get worried about?
- Some tests are more important than others. Ask yourself what would be the consequences of me filing this one, versus taking action?
- For some tests (such as potassium), we would usually follow up; but for haematocrit we probably wouldn't. One is very important and dangerous, the other less so.

Reading the result

All blood test results will have a similar layout and should contain:

- The patient or client identification code or number
- The person who requested the blood
- The test or investigation (take care with this one, as abbreviations and full biochemical names are often used interchangeably)

- Reference range
- Units
- Result.

We will explore each blood test in more detail throughout the book. However, at this stage it is worth looking at the units and range in more detail.

To read the result, first look at the reference range (more about that later) which has a small number and a larger number. These two numbers simply refer to the lowest and highest values expected in a normal population. The actual result is then usually presented next to the range. If the result sits outside the range, it is flagged up as 'out of range'.

For example:

- If the range is 6–20, and the result is 18, this is 'in range'.
- If the range is 6–20, and the result is 22, this is 'out of range' and will be flagged up using a red box, a star, bold font, arrows, 'H' for high (or 'low') etc, depending on the machine used.

Units

Be honest. Who looks at units? They can, however, provide some useful context when looking at a blood test result. The actual number could in reality be very big or very small and so it is often adapted so that it fits on the results report. To do this, we use the 'SI' system.

Here are a few examples:

- If we see x 10*9 used in reference to the white blood cell count (WBC), this means that the result has 9 zeros after it, so it's a pretty big number! A neutrophil result of 5 x 10*9/L actually means 5,000,000,000 cells per litre. Red blood cells are reported x 10*12/L; how many zeros do we have here? And do we have more red cells or white cells in the blood? The answer is red, by a ratio of 1000 RBC to every 1 WBC.
- If we see fL (femto), this means it's a very small number. The fL is often used to describe the size of the average red blood cell, mean cell volume or MCV. So, 85 fL is really 0.000,000,000,000,085, which makes sense because we know that our blood contains loads of red blood cells (x 10*12) but individually they are tiny.
- If we look at the renal or kidney markers, the blood test creatinine (usually 44–80) appears to be a bigger number than urea (usually 3–8). However, the former is micromol (u), which means it has 6 zeros and the latter is millimol (m) with only 3. A creatinine of 60 would be 0.000,060 whereas a urea of 6 would be 0.006. We therefore have much more urea in the blood than creatinine, despite urea initially looking like a much smaller number on the report.

- Some other units of interest are iU/L (which is used to measure an enzyme) and g/L (which is used to measure albumin). This type of measurement might be used in home cookery. For instance, if you saw a recipe for 40g sugar in 1 litre of water, you would need to use scales and a measuring jug. So there is clearly a lot of albumin in our blood! It transports other substances around our bodies and it has given us the terms 'corrected' or 'adjusted'. It also keeps water in the blood. As albumin levels decrease in the blood, the water is no longer retained by it and the albumin leaks out into surrounding tissue, causing swelling.

Clinical implications of results and understanding reference ranges

Blood tests are placed in context by reference ranges. Patients often fall outside these ranges, yet there is little or no clinical intervention. In this section, we look at how some of these test results can be affected by blood collection methods. We also discuss the ways in which reference ranges are constructed and used.

Blood collection and storage techniques

Tourniquets are often used to collect blood samples because they block venous return and cause dilatation, thus enabling easier identification of entry points. However, this typically causes loss of water and electrolytes from plasma, which may increase plasma protein levels. The stasis of blood flow can also produce different metabolic products (such as lactate); and if the patient is asked to clench their fist this may cause an artifactual hyperkalaemia (an elevated potassium level). These disadvantages do not mean that we should not use tourniquets. However, if results appear unreliable or unlikely – based on a patient's symptoms – it may be due to these factors.

Other problems include poor patient identification, samples taking more than 72 hours to be transported to the service, and samples being stored at the wrong temperature or not protected from light. To reduce the risk of these errors, each test has its own blood collection protocol.

The difference between plasma and serum

Plasma is the liquid component of the blood. It is predominantly made up of water, but also contains electrolytes and some proteins, glucose, hormones, CO_2 and the cells that make up the whole blood sample. Serum is the cell-free liquid component of the blood after clotting has occurred; thus fibrinogen, cells and other clotting factors are not present. Serum is usually obtained by drawing whole blood into a gold-top SST tube, or a red-top tube, which will

induce a clot, from which the liquid serum can be isolated. It will still contain electrolytes, antibodies and hormones.

Serum is actually the Latin word for 'whey' (as in 'curds and whey', the products formed when milk is allowed to curdle). The whey is the liquid component and the curds are the solid parts. If blood is allowed to clot, the liquid component is therefore called serum. If blood is prevented from clotting, the liquid component is called plasma.

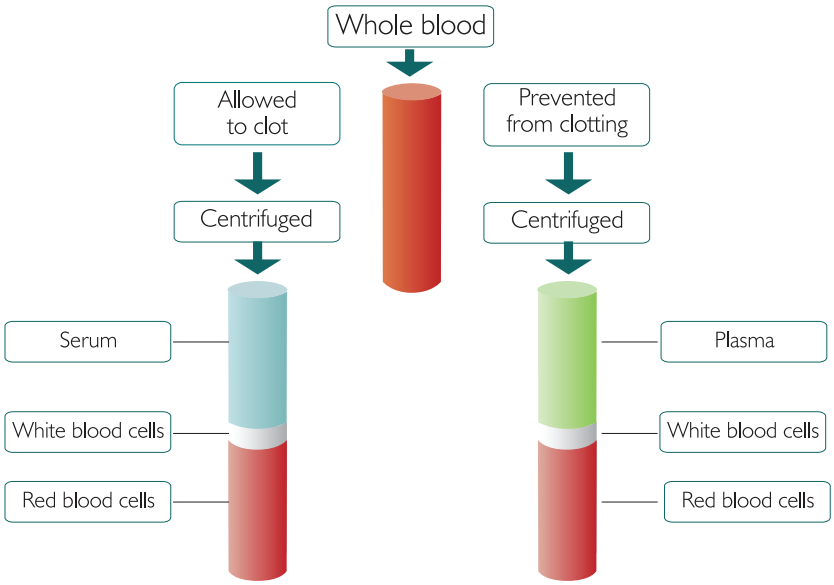


Figure 1.1: Composition of whole blood, serum and plasma

The main disadvantage of using serum is that the blood has already clotted. In other words, a series of metabolic processes occurred after the sample was collected but before the sample was measured. This can lead to errors in measuring such elements as potassium, phosphate, magnesium, aspartate aminotransferase, lactate dehydrogenase, serotonin, neurone-specific enolase and zinc content. The advantage is that serum can be used to measure constituents that would be destroyed or compromised by the anticoagulant chemicals used in the preparation of plasma samples such as serum protein electrophoresis (SPEP).

The main disadvantage of using plasma (blood that has not clotted, due to the addition of anticoagulants) is that the anticoagulants can interfere with certain analytical methods or change the concentration of the constituents being measured. The advantages of using plasma samples include 'cleaner' samples that have not undergone the clotting process, time saving and a higher yield (up to 20%).

Different types of anticoagulants are denoted by different-coloured caps on collection tubes. Typical anticoagulants include sodium heparin (green), sodium fluoride (grey), sodium citrate (blue) and EDTA (lavender). Your local healthcare setting will have a tube guide showing the active component of the tube. A clear cap means that neither a clot-activating (serum) nor an anticoagulant (plasma) is present; these clear-capped tubes are often used as discard tubes. The order of draw can be important if multiple tubes are used, to avoid contamination from one tube to the next. Check with your local healthcare setting, but a typical order of draw using the vacutainer system would be:

1. Blood culture
2. Sodium citrate (light blue) for coagulation testing (PT, INR, aPTT) and D-Dimer
3. Serum (red) for LDH, ionised Ca, drugs (phenytoin, theophylline, methotrexate, lithium), vitamin D, parathyroid hormone, osmolality, bone markers, endocrine testing (excluding thyroid)
4. Serum separator tube (SST) (gold) for thyroid function (TSH, FT4, T3, cortisol), nutritional markers (gastrin, B_{12} folate, ferritin), tumour markers (PSA, CEA, AFP, HCG, CA125, CA19.9, CA15.3), immunoglobulins (IgG, IgA, IgM, IgE), electrophoresis, CRP, thyroid Ab, liver Ab, rheumatology Ab
5. Plasma separator tube (PST) (light green) for U&Es, LFTs, cardiac enzymes, Ca, Mg, phosphate, uric acid, total protein, amylase, lipids, bone profile, troponin, iron status
6. EDTA (lavender) for full blood count (FBC) and ESR, haemoglobin A1c, homocysteine (send on ice and state time), ACTH
7. Cross match (pink) for blood transfusion samples
8. Fluoride oxalate (grey) for glucose
9. Trace element (royal blue) for lithium and magnesium

Haemolysed samples

If, following centrifugation, the plasma or serum looks reddish rather than straw yellow, the sample has probably haemolysed. In a haemolysed sample, some of the red blood cells have lysed (broken open) and their contents will have contaminated the plasma or serum sample. This will cause errors in reporting, for example, elevated potassium, magnesium and phosphate. The laboratory may be able to negate the effect of using a haemolysed sample if the result is needed urgently or it is difficult to obtain another sample. Common reasons for the sample being haemolysed include:

- the collection needle gauge being too narrow
- over-vigorous shaking of the sample

- an underlying haematological disorder
- red cells being isolated for storage and then stored in water or a non-isotonic solution
- over-vigorous dispensing of blood from the hypodermic syringe to collection tubes.

Reference ranges

Most people are comfortable with the idea of reference ranges, but what do they actually tell us? Or, rather, what *don't* they tell us? To create a reference range, a number of volunteers (usually over 120) are matched for factors such as age, gender and ethnicity, and the analyte of interest is then measured. Firstly, most ranges have a 95% confidence, which means that the top 2.5% of values and the bottom 2.5% of values are omitted. In other words, it is possible to be healthy but outside the reference range because you are at the very top or the very bottom, neither of which are shown. Secondly, you should use ranges from unvalidated sources with great care, as ranges can vary with age and gender and local population. Best practice is to use the range that is presented with the actual value.

Storytelling: If you measured the height of 120 shoppers at a supermarket, the world's smallest man and the world's tallest man might be present and be included in your sample. However, it's unlikely that you'll see them again, so the top and bottom of the data is cropped, leaving more of the average making up the range. This is the first problem with reference ranges. Of the healthy cohort, around 5% are excluded at this initial stage, leaving them outside the range.

Range hangover

As we have seen, reference ranges refer to us on 'good day' or peak health. However, most of our patients will be 'out of range' so we need a strategy to deal with this. Firstly, let's consider the idea of range hangover.

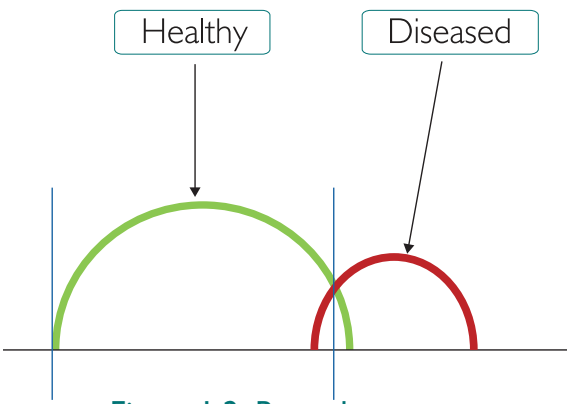


Figure 1.2: Range hangover

In Figure 1.2 (above), the x-axis shows the increase in number from left to right. The green line represents a healthy (usually asymptomatic) individual and the red line a patient with a disease (usually symptomatic). The two blue vertical lines represent the reference range. The line on the left is the lowest value and the one on the right is the highest. Because the upper range blue vertical has been drawn in a particular place (called the cut-off value), some of the green line 'overhangs'. This means that the patient who is healthy may be reported 'out of range'.

Clinical sensitivity and specificity

Because most blood tests have an associated pathology but may not be very accurate in detecting a specific disease, they sometimes produce 'false positives'. Clinical specificity refers to whether the test can correctly report someone without the disease as being 'healthy'. Conversely, clinical sensitivity refers to whether the test can correctly report someone with the disease as being 'diseased'.

The terms used to describe this are:

- True negative (TN): patient is healthy and blood test result is within reference range
- True positive (TP): patient has condition and blood test result is outside the reference range
- False negative (FN): patient has condition but the blood test result falls within the normal reference range
- False positive (FP): patient is healthy but the blood test result is outside the reference range.

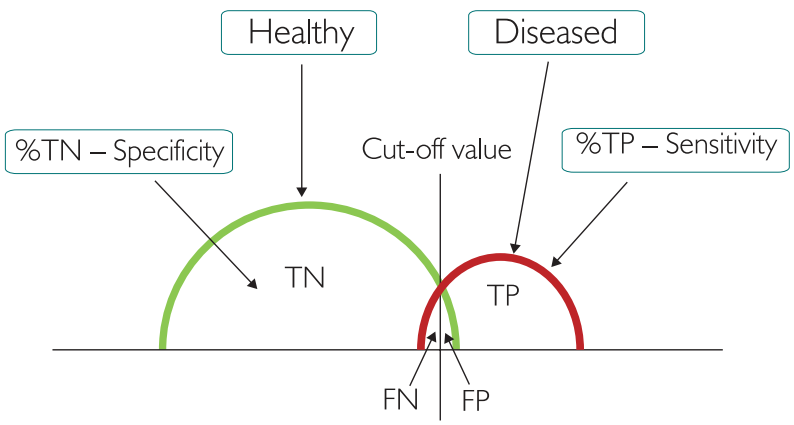


Figure 1.3: The cut-off value between 'healthy' and 'diseased'

The cut-off value is the point at which people change from being labelled 'healthy' to being labelled 'diseased', or the reverse. If we move the cut-off value to the far right, everyone

who is healthy will be reported as healthy, but more diseased people will be missed (because they are wrongly labelled healthy). If we move the cut-off value to the far left, everyone who is diseased will be reported as diseased, but more healthy people will be wrongly labelled 'diseased'. At this point, we need to consider factors such as cost of screening, reliability of data and (most importantly) medical ethics. Is telling someone they have cancer when they don't as serious as missing someone who *does* have cancer?

Clinical decision limits (CDLs)

A clinical decision limit (CDL) allows us to prepare protocols and flow charts and consider patient journey management which often informs what we do next.

If we look again at the range hangover diagram (Figure 1.2) but this time add several arbitrary additional cut-off values, we can give each of these a letter and assign typical conditions and actions to each one. (Remember to seek local confirmation as these are only examples to illustrate the theory.)

In Figure 1.3 (below) the disease group is divided into sections A to D, based on the values being 2-fold, 4-fold and 10-fold higher than the upper range.

In practice, if we consider the liver enzyme alanine aminotransferase (ALT) this usually has an upper limit of 40. Our CDLs may therefore look as follows:

- 'Normal' reference range for ALT = (10–40)
- Group A = a blood test result for ALT between 40 and 80 (2x the upper normal limit of 40)
- Group B = a blood test result for ALT between 80 and 160 (4x the upper normal limit of 40)
- Group C = a blood test result for ALT between 160 and 400 (10x the upper normal limit of 40)
- Group D = a blood test result for ALT over 400 (greater than 10x the upper normal limit of 40)

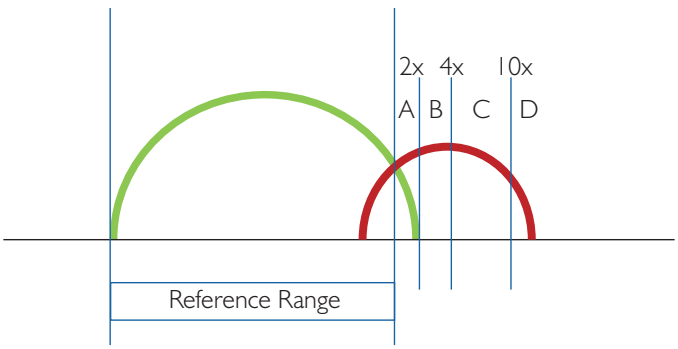


Figure 1.3: The cut-off value between 'healthy' and 'diseased'

Strategy for values outside the range

Quite often the patient's blood test results will fall outside the reference range. As blood test results take the form of numbers and are not binary (like a broken arm), they can be viewed subjectively. It may be helpful to ask yourself the following questions:

1. How close is the value to the limits of the reference range? Consider FN, FP, TN, TP, and the mathematical limitations of reference ranges discussed above. This point is about the inherent variance in the range, person, blood sample, and so on.
2. Has the test been repeated?
3. Is the value increasing or decreasing over time?
4. Has the patient had surgery or intervention? An elevated range of inflammatory markers, such as C-reactive protein (CRP), erythrocyte sedimentation rate (ESR) and interleukin 6 (IL6) would be expected post-surgery.
5. Does the blood test fit with other 'indices' or family markers? Most tests fit into a family group (Hb, RBC and Hct). This point will be further explored in the case study in Chapter 4.
6. Is it clinically relevant? The patient may have competing pathologies. Some values will therefore be outside the range because they represent a pathology or condition that is being managed, such as chronic obstructive pulmonary disease (COPD), alcoholism or diabetes. You may be looking for a new out-of-range value or an unusually high value.
7. Will it change the patient's treatment? Before requesting the test, consider what you will do with the result and why you are requesting it.

However, you should bear in mind that the above questions should always be used in conjunction with local clinical procedures in your own healthcare setting.

We may therefore choose to do the following by applying the four key questions we outlined earlier:

Question 1: How far out of range is the result?

Question 2: Does it make sense? Is the patient symptomatic?

Question 3: Do the other family group blood tests agree?

Question 4: Is this an important blood test?

You may choose to construct a decision table.

I have made a start on one below.

Example clinical decision table for the liver enzyme ALT:

Group	**Q1 – How far out of range?**	**Q2 – Does it make sense?**	**Q3 – Family groups?**	**Q4 – Important blood test?**	**Possible action**
A	Not far out of range – contains the green false positive overhangs	No symptoms	All other liver function tests are in range	Yes	Consider filing?
A	Not far out of range	No symptoms but recently started on a new medication	All other liver function tests are in range	Yes	Repeat bloods?
A	Not far out of range	Symptomatic	Yes, other markers are also out of range	Yes	Repeat bloods? Consider referral or scan?
D	Significantly out of range	Very likely to be unwell and symptomatic	Other markers are very likely to be out of range	Yes	Urgent action?

We can do this with any blood tests. Let's try C-reactive protein, which increases with more cellular damage (inflammation). This one is a little trickier, given the timing of CRP and the white cell response (see Chapters 7, 8 and 11).

Example clinical decision table for the inflammatory marker CRP:

Group	**Likely condition?**	**Does it make sense?**	**Family groups?**	**Important?**	**Possible action?**
Normal (3–10)	Normal – could this be a false negative, as is often seen in rheumatoid arthritis (RA)?	No symptoms	All in range (white blood cells)	Yes	File?
A: 10–20 (2x)	Autoimmune, CVD	Symptoms of RA present	Yes, WBC and neutrophils and globulins	Yes	File – normal for a diagnosed RA?
B: 20–40 (4x)	Viral	Symptoms	Yes, WBC and lymphocyte and globulins	Yes	Consider a virus?
C: 40–200 (10x)	Bacterial infection	Symptoms	Yes, WBC and neutrophils	Yes	Action required
D: (200+)	Trauma/Sepsis	Yes – recently had an operation = trauma			
No = bacterial	Yes, WBC and neutrophils	Yes	Red flag		

There are lots of examples of CDLs being used, from tracking troponin levels in heart attacks to the level of creatinine kinase (CK) in rhabdomyolysis. We then have the extra challenge from the four questions, linked to family groups. We need to ask what we are actually measuring, in order to propose likely actions.

Please note that in these examples we have looked at a pathology which increases blood test values. In patterns of anaemia, for example, the cut-off groups A–D would be less than normal.

Self-assessment

1. Name two components of the blood. Can you give an example of each type?

2. List four ways in which the blood can change, using words that make sense to you. Can you list some examples for each type of change?

3. What are the four questions you need to consider when reviewing a blood test?

4. Using symptoms and blood test results, define 'false positive' and 'false negative'. Can you give any examples?

Quick reference glossary

The following table shows common terms, abbreviations and some typical observations relating to the various blood tests. Some examples also have metaphors, shown in quotation marks, to aid memory and understanding. These will be explained further in the corresponding chapters.

Table 1.1: Glossary of terms used in blood tests

Full blood count (FBC) – measures haematology of cells	
Platelet	• Cell that causes the blood to clot • Also a marker of bone marrow function • Decreased in some leukaemia and myelomas • Additional test is mean platelet volume (MPV) • Low (usually less than 150 10*9/L) is called thrombocytopenia; platelets are decreased in bleeding, liver disease, pregnancy, infection and due to medication • Raised (usually greater than 450 10*9/L) is called thrombocytosis; platelets are raised in infection, inflammation, trauma, post operation or due to medication
White blood cell count (WBC)	• The total number of white cells in the blood

Neutrophil	• A type of white blood cell • Responds to tissue damage via C-reactive protein (or CRP) • Raised in bacterial infections, autoimmune conditions • **'The fire engine'**
Lymphocyte	• A type of white blood cell • Makes antibodies • Raised in viral infections and some myelomas • **'The police'**
Monocyte	• A type of white blood cell • Infiltrates the tissue in systemic bacterial infections • Linked to cardiovascular disease and high low-density lipoprotein (LDL) cholesterol • **'The miner'**
Basophil	• A type of white blood cell • Important in allergic responses and hypersensitivity
Eosinophil	• A type of white blood cell • Important in allergic responses and hypersensitivity
Blast/Atypical	• A type of dysfunctional white cell • Raised in leukaemia and myelomas
Haematocrit (Hct)	• Percentage of red blood cells in the whole blood • Decreased in anaemia • Elevated in polycythaemia
Haemoglobin (Hb)	• The oxygen-carrying protein in the red blood cell • Decreased in anaemia • Elevated in polycythaemia
Red blood cell count (RBC)	• The total number of red blood cells in the blood as a count • Decreased in anaemia • Elevated in polycythaemia

Mean cell volume (MCV)	• The average size of the red blood cells • Low in iron deficient anaemia • Normal in blood loss anaemia • High in folate and B_{12} deficient anaemia
Inflammatory markers – measures biochemistry	
Plasma viscosity (PV)	• A measure of more 'stuff' in the blood • Thus, a surrogate, non-specific marker of inflammation • Increased in autoimmune conditions, infection, cell damage, cancer, myelomas • **'The traffic jam due to fire engines and police (white cells)'** • Could remain raised for two weeks post-injury, as increased white cells have around two-week lifespan
Erythrocyte sedimentation rate (ESR)	• How quickly red blood cells fall in a tube, in a lab • A surrogate, non-specific, marker of inflammation that has elicited a fibrinogen response • Fibrinogen 'sticks' red blood cells together so they become heavier and fall more quickly • Could be normal in low damage inflammation as seen in some autoimmune conditions • **'The scaffolding and building-supporting structure following a large fire'** • If raised, could remain raised for a significant time post-event
C-reactive protein (CRP)	• A chemo-attractant protein released in response to tissue damage • **'The fire alarm'** • Possible to miss the CRP response post-injury whilst still having raised PV and ESR • Increasingly being used as a sensitive marker for atherosclerotic vascular damage to indicate cardiovascular risk

Urea and electrolytes (U&Es), Kidney function – measures biochemistry	
Sodium (Na)	• Extracellular electrolyte that controls water balance and blood pressure • Raised in dehydration, ◀▶ urea
Potassium (K)	• Intracellular electrolyte, controls cellular pumps and receptors via electric potential • Therefore a red flag if in high concentrations in the blood
Urea	• A marker of acute renal dysfunction, such as distress, although this could be something like dehydration, so ◀▶ to Na levels
Creatinine	• A marker of chronic renal function, such as a renal stone
Estimated glomerular filtration rate (eGFR)	• A general marker of kidney function • Used to diagnose chronic kidney disease staging • Used to confirm renal dysfunction as cause of other conditions such as renal anaemia
Liver function tests (LFTs)– measure biochemistry	
Alanine aminotransferase (ALT)	• A liver enzyme • Often raised in trauma, drug toxicity, and viral hepatitis
Aspartate aminotransferase (AST)	• A liver enzyme • Often raised in trauma, acute alcohol hepatitis and liver failure • Also found in the heart so ◀▶ to cardiac markers/ chest pain
Gamma-glutamyl transferase (GGT)	• A liver enzyme • Often raised following alcohol intake • ◀▶ to RBC, MCV and folate to differentiate between alcohol, B_{12} and diabetes neuropathies
Alkaline phosphatase	• A liver enzyme • Often increased in biliary tree damage such as gallstones Also found in the bone (check Ca), kidney (check U&Es) and placenta (check age and gender)

Amylase	• A liver enzyme • Often increased in pancreatitis and pancreatic tumours
Bilirubin	• A marker of the 'plumbing' of the liver • Increased in jaundice, usually caused by pre-, actual or post-hepatic blockage
Urobilinogen	• A bilirubin breakdown product, usually absent in post-hepatic jaundice
Albumin	• A protein produced by the liver • A chaperone for chemicals like Ca so could give false low value in nutrient-deficient patients • Decreased in liver damage
Globulin	• A crude marker of antibody production/presence • Often increased in autoimmune conditions, myelomas and viral infection
Additional tests – measure biochemistry	
D-Dimer	• A breakdown product of a clot • Care should be taken to link D-dimers to deep vein thrombosis (DVT) and pulmonary embolism (PE) • Refer to NICE guidelines (UK)
International normalised ratio (INR)	• This is a marker of blood clotting • How long it takes for your blood to clot is given a baseline value of 1, thus an INR of 2 would mean your blood is taking twice as long to clot • Goes up in anti-coagulation and liver disease
Bence Jones protein	• A breakdown product of a 'nonsense' antibody • Usually present in a myeloma
Bone profile	• Usually returns corrected Ca, PTH and vitamin D_3 • Can help differentiate between osteomalacia (rickets), Paget's and osteoporosis

Prostate specific antigen (PSA)	• Released by the prostate • Relative to prostate damage • A slightly raised PSA may not mean prostate cancer • Link to urea and Alk phos, and Ca (secondary bone metastasis)
CA-125	• A marker of ovarian cancer
Thyroid function	• Thyroxine (T4), thyrotrophic releasing hormone (TRH) and thyroid stimulating hormone (TSH) are measured to diagnose cause of primary, secondary or tertiary hypothyroidism (or hyperthyroidism); additional confirmatory tests may be required • Also used to titre T4 supplements
Autoimmune markers	• Rheumatoid factor (rheumatoid arthritis) and ankylosing spondylitis (HLAB27) are types of self-antibodies that are often present in autoimmune conditions • Intrinsic factor and parietal cell antibody for pernicious anaemia
Haemoglobin with glucose irreversibly bound (HbA1c)	• A long-term marker of glucose in excess, used in diabetes monitoring
Acid Base pH Bicarbonate	• Used to monitor respiratory (chronic obstructive pulmonary disease) and metabolic (drug overdose) acidosis or alkalosis
Ferritin (Iron), Folate, B_{12}	• Nutrient markers to be used with RBC, Hb and MCV • Low MCV usually has low ferritin (high ferritin in hemochromatosis) • High MCV usually has low folate and/or low B_{12} • Low folate from sustained alcohol, drug interactions, diet or gastro-intestinal (GI) conditions • Low B_{12} from GI conditions, diet and autoimmune pernicious anaemia

Troponins, creatine kinase mb (CKmb), B-type natriuretic peptide (BNP)	• Cardiac event markers • Troponins and CKmb are proteins found in the cardiac tissue that are present in high concentrations in the blood following a cardiac event • BNP is a peptide found in the cardiac wall; increased levels may mean ventricular wall load is dysfunctional and may predispose to a cardiac event

2

What are we actually measuring and why does this matter?

As we outlined in Chapter 1, we usually measure a cell or chemical in a blood test so it's important to define what we are actually measuring. In this chapter we will look at the four key ways in which blood tests can change. Once you've read this chapter, re-read Chapter 1, especially the section on clinical decision limits, in the light of the information from this chapter. In Chapter 2, we will also outline some specific examples of applied blood tests (such as the liver enzyme ALT). You will find out more about each of these blood tests in their respective stand-alone chapters.

Part 1 – Cellular content

In this example, the substance we are measuring should be found inside cells or organs doing its job. It shouldn't be found in large amounts in the blood. The main way these chemicals are found in the blood is if they have 'leaked out' of the host cell due to cellular damage.

Storytelling: Think about a typical car engine, which contains engine oil. One day you look on the driveway under the car and see no oil, which is a good thing! The oil should not be on the driveway; it should be in the engine, doing its job of lubricating the engine. Another day, you notice a small amount of oil on the driveway, directly under the engine. Oh dear. The oil has probably leaked out of the engine. You can judge the severity of the problem by noting how much oil is on the floor. This is similar to our earlier discussion about clinical decision limits.

Examples of cell leakage

Liver enzymes alanine aminotransferase (ALT) and aspartate aminotransferase (AST) should both be inside the liver doing their jobs. If the liver cells get damaged (for example, by medication, viral infection, alcoholism, or paracetamol overdose), these enzymes leak out of the damaged cells and we start to see them in the blood. This is why we often track ALT in patients with prescribed medication, as it is a marker of liver cytotoxicity. A very slightly raised ALT (Group A up to 2x the upper limit) is common and could simply be due to normal levels

of liver regeneration (the ALT will still leak out). Values less than 80 are therefore often filed if no symptoms are present.

The muscle enzyme creatinine kinase (CK) is found inside the muscle. As the muscle is damaged (e.g. by dehydration, muscle injury, statin damage or malnutrition), CK leaks from the muscle into the blood, with 10x the upper limit associated with rhabdomyolysis.

Potassium (K) is found inside muscle cells and can leak out if muscles are damaged. It is also found in most cells and will leak out if a cell is damaged, or in an acidic environment where H+ enters the cell and K+ is pushed out into the blood. It is also found in red blood cells and platelets and can become artificially raised in a blood tube, if the blood is taken or stored correctly.

Troponins are specialist proteins found inside cardiac cells. As these cells are damaged (usually by hypoxia), the troponin leaks out and we can therefore map troponin levels in the blood reflecting cardiac damage over time. Brain natriuretic peptide (BNP) is used in a similar manner, as it leaks out when the cardiac wall becomes damaged and is often used to predict a cardiac event.

Note: These blood tests only work if the original organ is healthy and allows for these chemicals to leak out. For example, a patient with heart failure may have lower than expected troponins. Likewise, a patient with prolonged liver damage caused by alcohol dependency may have lower than expected liver markers because they have a very small viable liver from which these chemicals can leak out. However, in both these examples, the patient will clearly be unwell and it will be helpful to refer back to 'Question 2 – Does the result make sense?' (discussed in Chapter 1).

Part 2 – Clearance of waste products

In this section we will talk about two waste removal systems, the kidney and the liver. In this example the blood test we are measuring is actually a waste product which should be removed from the blood by the kidney or the liver. If these organs are not working properly, the amount of these waste products will increase in the blood (as they are not being removed into the urine or faeces).

Examples of waste products removed by the kidney

Urea in the blood is cleared by the kidney into the urine. If the kidney is not working properly, levels of urea will usually rise in the blood. Increasing urea in the blood suggests that the kidney is not working properly and therefore won't be able to effectively regulate electrolytes like sodium and potassium. This could lead to a 'red flag' event, such as a cardiac arrest.

Creatinine is also removed by the kidneys. However, as we have less creatinine, creatinine levels take longer to change noticeably. If creatinine levels increase rapidly over 5–7 days, this may therefore be recorded as 'Acute Kidney Injury' (AKI) and we may then urgently review the patient's medicines, for example.

We also use creatinine to measure how good the kidney is at filtration. This result is called the glomerular filtration rate (GFR). We don't usually have the body mass index (BMI) and other information needed to calculate it exactly so it's normally called eGFR, with the 'e' standing for 'estimated'. Most people aged 18–35 have an eGFR of around 100mL/min. This means that their kidney is filtering 100mL of blood per minute, which is pretty good going. (If you're ever having a challenging day, at least you can thank your kidneys for all that lovely filtering they are doing for you!) However, once we reach the age of 35, our eGFR usually drops by about 1% per year. So it is usual for someone who is 90 years old to have an eGFR of 30–40mL/min. Of course, some 90-year-olds have an eGFR of 100 (just as some 90-year-olds run the London marathon) but they are the exception.

Let's consider eGFR in more detail. If we are expecting an eGFR of 100mL/min but our patient has an eGFR of 25mL/min, their kidneys will take four times longer to filter or process the blood. This could have significant effects on prescribing drugs which are cleared or processed by the kidney and we may therefore need to adjust the dosage and times accordingly. The eGFR can also be affected by ethnicity – check local protocols for more information.

Other examples of waste products not being cleared and therefore increasing in the blood can be uric acid (in the case of gout) and some false positive markers for myeloma.

Examples of waste products removed by the liver

The waste product removed by the liver that we measure in the blood is called bilirubin. Bilirubin is found inside red blood cells and leaks out when the red blood cell 'dies', at the end of its lifespan, and is broken down. When a bilirubin molecule is removed from a red blood cell it is called 'indirect' or 'unconjugated' bilirubin and this part of the liver system is called 'pre-hepatic'. The bilirubin is then modified in the liver by joining it with another bilirubin molecule to form 'direct' or 'conjugated' bilirubin. This form is then excreted via the biliary tree (through the gall bladder and so on). This part of the liver system is called 'post hepatic' and the cells here contain large quantities of a liver enzyme called alkaline phosphatase (ALK or ALP). Increased amounts of bilirubin in the blood are present in patients with jaundice.

We often report total bilirubin, which is a sum of direct and indirect bilirubin, and so we need to look at family groups to see which type of jaundice the patient has.

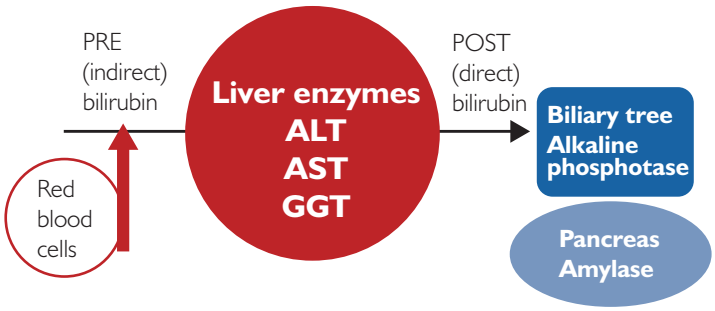


Figure 2.1: Liver function

In Figure 2.1 (above), we could review LFT jaundice as follows:

Table 2.1: Sample LFT jaundice results

Patient	RBC	Bilirubin	ALT	Albumin	ALP	Jaundice?
A	High	High	Normal	Normal	Normal	Yes: Pre-hepatic
B	Normal	High	Normal	Normal	High	Yes: Post-hepatic
C	Normal	Normal	Slightly raised	Normal	Normal	No
D	Normal	Very high	Very high	Low	Very high	Yes: Post-hepatic with actual liver damage

Part 3 – Production

In this example we will look in detail at how the blood results change in response to increased or decreased production. Typical pathologies here include 'inflammation', 'infection', 'clotting', 'bleeds' and patterns of 'anaemia'.

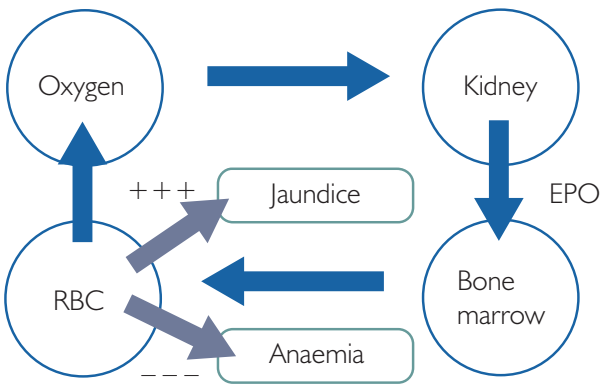


Figure 2.2: EPO production

Red cell production

Red blood cells are produced by the bone marrow and spleen under the action of a hormone called erythropoietin (EPO), which is produced by the kidney. If the kidney is not working properly, it may not be able to produce EPO and red cell production may therefore be compromised. This is called renal anaemia.

Looking at Figure 2.2 (above), the kidney produces EPO, which acts on the bone marrow to produce more red blood cells. If the patient produces too many red blood cells, they may make too much bilirubin and this may lead to a pre-hepatic jaundice, as discussed earlier. If the kidney is unable to make EPO, the patient may make fewer red blood cells and develop anaemia. This process is often called 'IN/OUT'. For example:

- Kidney: Oxygen/CO2 levels IN – EPO OUT
- Bone marrow: EPO IN leads to RBC OUT
- Liver: Indirect bilirubin IN – direct bilirubin OUT

You may find it helpful to draw a network diagram with the key organs and connect them with IN/OUT arrows, as in Figure 2.3 (below). You can then run scenarios by switching off the kidney and seeing which IN/OUT arrows would be changed. This can help you map interconnected symptoms to the blood tests.

White cell production

We have a similar IN/OUT diagram for white cell production. In Figure 2.3 (below) cell damage releases a chemical called TNF, which goes to the liver. This in turn produces CRP, which induces the liver to make more neutrophils and thus more white blood cells.

Liver – IN (TNF) and OUT (CRP)
Bone marrow – IN (CRP) and OUT (Neutrophils/WBC)

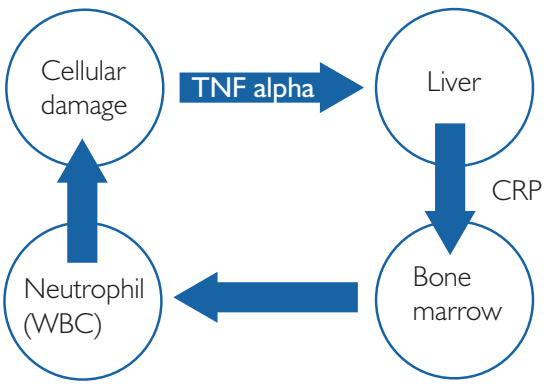


Figure 2.3: White cell production

Storytelling: We can consider this pathway as the response to a fire (cell damage), which produces a fire alarm (TNF). CRP then causes the fire engines to be sent to the scene.

This response is summarised in Figure 2.4 (below).

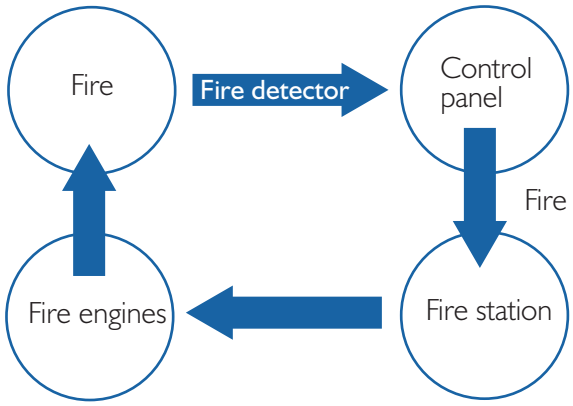


Figure 2.4: Response to cell damage

This can help explain the time lag between the CRP response (immediate) and the WBC response which may occur a few days later. Given that white blood cells have a lifespan of 3–4 weeks you may see the following:

- Scenario 1: High CRP, Normal WBC (neutrophil) = Acute. The WBC response is yet to happen.
- Scenario 2: High CRP, High WBC (neutrophil) = Acute, WBC has responded.
- Scenario 3: Normal CRP, High WBC (neutrophil) = An event probably happened over the last 4 weeks; may not be acute now.
- Scenario 4: Symptoms of inflammation but normal CRP and neutrophils (false negatives). Could this be due to a liver or bone marrow problem? Could this be a localised symptomatic response which has not reached the liver, perhaps due to poor vascular flow in the area (e.g. a degenerated knee)?

Scenario 4 is explained in Figure 2.5 (below) in which the local response to swelling (symptoms) and the systemic blood tests are highlighted.

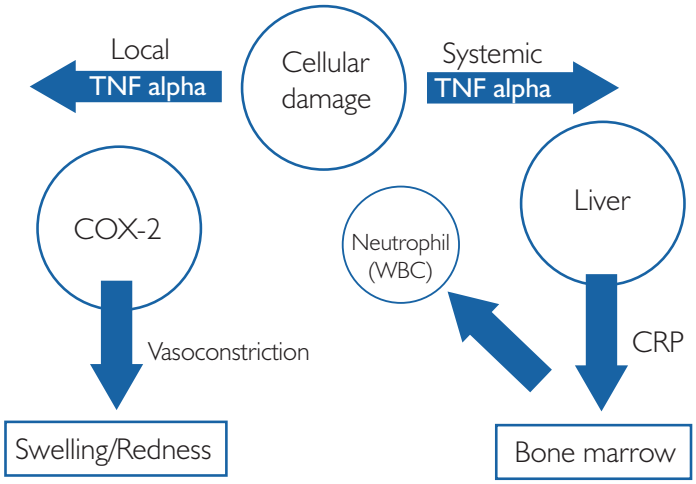


Figure 2.5: Alternative pathway – cell versus exudative

This pathway is explored in more detail in Chapter 7 (on inflammatory markers).

Part 4 – Interaction

In the final section we will look at interaction. This is when more than one system is responsible for the blood test we are measuring. Two examples are albumin and HBA1C.

Albumin is produced by the liver and kept in the blood by the kidney. If either organ fails, albumin levels can fall.

HBA1C really means haemoglobin with glucose stuck to it. Glucose will stick to anything, forever. And we can easily measure red blood cells because we can look to see how much glucose is stuck to them. If they have lots of glucose stuck to their red blood cells, the patient will probably also have lots of glucose stuck to their nerves (diabetic neuropathy) or to their kidneys, or to their eyes (retinopathy). Because red blood cells have a three-month lifespan, the HBA1C test reveals patient compliance over three months. It's not possible to cheat this test by behaving well for a week before a diabetes review! On the other hand, it's not good at diagnosing rapid onset diabetes, in pregnancy or in people with severe anaemia (as the test requires red blood cells in order to work).

3

Family groups

In this chapter we will look at family groups. Remember that this is one of the four questions we talked about earlier, in Chapter 1. I use the term 'family group' to describe a group of blood tests that are linked together. As with witness statements in a court case, it helps when all the blood tests in a family group provide similar information – such as all being 'out of range' or all being 'in range'. You may want to call them associated tests, linked tests or even 'this blood test's mates'! All are good.

Grouping blood tests together can be very effective. Group results are great for ruling stuff in, and ruling stuff out. They may also suggest additional tests that you may want to request.

I'd like to challenge you to try something different when reading the blood results. Rather than looking at the red 'out of range' stuff first and trying to fit a story to it, try using differential diagnosis, in which you rule things in and out. This is helpful for two reasons. Firstly, remember those false negatives from earlier? Using a family group could help you identify them. Secondly, we sometimes get some strange results. For example, you may have requested a 'liver function test' and some of the results are out of range, but it turns out that the liver is completely fine. This is a case of 'masquerading'.

Full blood count

Let's start with the full blood count (FBC) test. The bone marrow manufactures three key cell types: red blood cells (erythrocytes), white blood cells (leukocytes) and platelets. This is our first family group:

1) Haemoglobin (Hb) – a marker of red blood cells

2) White blood cells (WBC)

3) Platelets (PLT)

This family group gives us information about the cells produced by the bone marrow. It may therefore give us helpful information about conditions such as leukaemia (raised WBC, low HB and low PLT) and/or myeloma, etc. This example involves one person with a single condition (leukaemia).

We could also have the same FBC profile in one person with three different problems: raised WBC from an infection, low platelets from infection and medication, and low red cell count from anaemia. Knowing the patient is clearly very important. Remember the question – does the result make sense?

Next time you look at a blood test report, try to find Hb first, then WBC and PLT.

Red blood cells

Let's now look in more detail at the red cell family – or, to use a fancy word, indices.

Hb has two 'mates' or friends. These are haematocrit (HCT) and red cell count (RCC), also known as red blood count (RBC). HCT describes what percentages of the whole blood are red blood cells. About 40–50% RBC describes the absolute amount of red blood cells (lots!) per litre and haemoglobin measures the amount of oxygen-carrying protein in the blood, most of which is inside red blood cells.

It may be helpful to sub-code these as: 1 (HB, from above), 1a (HCT) and 1b (RBC) to show that these are linked.

If all three red cell markers/family/indices are low, this is called anaemia and we would then look at mean cell volume (MCV) which we could code as 1c (MCV). We may then decide to request further tests to explore anaemia, such as 1d (ferritin), 1e (B12) and 1f (folate). More detail on this can be found in Chapter 5 (on the patterns of anaemia).

We also use the red cell groups to look for false positives. Look at this blood result:

1) Hb: Normal

2) RBC: Normal

3) HCT: Slightly decreased (in the A group, as discussed earlier)

In this example, the fact that Hb and RBC are both normal would suggest (assuming the patient is asymptomatic) that the HCT result is a false positive and not clinically significant. Its 'mates' don't back its story up. It's not a very important marker, so a protocol would probably suggest that we note, and then file it.

White blood cells

Next, let's look at the white cell family. WBC is the total amount of white blood cells. If that is 'out of range', we need to know which type of white cell is elevated and has increased the overall total WBC count.

There are several types of white blood cells. The most common subtype is the neutrophil. This usually accounts for around 75% of our white blood cells. Look at the units to see which

ones you are reading. Usually the count (x10*9) is more accurate. The percentage is often called 'the white cell differential' and can help show if we have switched production to a different type of white blood cells. Neutrophils respond to C-reactive protein (CRP) so this may be an additional test to request, or to look for (if you already have it).

The next type of white blood cell is the lymphocyte, which accounts for between 25 and 30% of the white blood cells. We then have monocytes, eosinophils and basophils making up the remaining 5 to 10%.

We could therefore sub-code these as: 2 (WBC), then 2a (neutrophils), 2b (lymphocytes), 2c (monocytes), 2d (eosinophils) and 2e (basophils).

Liver function tests (LFTs)

Depending on the setting, a typical common LFT would be:

1) ALT

2) Albumin (ALB)

3) Bilirubin

4) Alkaline Phosphatase (ALP)

Thinking about the section describing how blood results change, we can see that these have been chosen to show:

1) ALT: Actual liver cell content – leaks out into blood when liver is damaged

2) ALB: Liver produces albumin (production)

3) Bilirubin: Waste product

4) Alkaline phosphatase: Found in biliary tree and post-hepatic obstructive jaundice (cellular content)

We can therefore use this family group to look at pre-post hepatic jaundice and masquerading co-morbidity.

For example:

1) ALT: Normal

2) Bilirubin: Normal

3) Albumin: Normal

4) Alkaline phosphatase: Increased

Because the ALT, bilirubin and albumin markers are in range (and assuming they are asymptomatic for liver), it is likely that raised ALP is not caused by the liver.

Alkaline phosphatase is found in the following organ systems:

1) Placenta

2) Liver

3) Kidney

4) Bone

We can use family groups to determine which organ or system is responsible for the raised ALP:

1) Placenta: OK, we don't need a blood test for this one!

2) Liver: Look at ALT and bilirubin

3) Kidney: Look at urea, (acute kidney injury markers and eGFR)

4) Bone: Look at bone profile – corrected calcium is a good marker

The LFT may also reflect your setting, so if you request an LFT in primary care you may get test results as follows:

1) ALT

2) ALB

3) ALP

4) Bilirubin

This makes sense, as these are early indicators that something is wrong.

However, if you work in a setting with more acutely unwell patients (which can still be in primary care), community, emergency, or with drug and alcohol clients, your LFT request may return the following blood test results:

1) ALT

2) ALB

3) ALP

4) Bilirubin

5) AST

6) GGT

AST and GGT are now included because of the nature of the cohort being tested and because these results are likely to be of additional benefit. There is a cost associated with all blood testing so only the salient tests should be requested. The cost does become an important consideration when scaled up across a large practice, hospital, region and country.

One final point to note here is that some enzymes are not exclusive to one organ system. We have seen, for instance, how alkaline phosphatase is found in multiple systems. Likewise, the liver enzyme AST is found in the heart tissue as well as the liver. If you have requested a liver function test (LFT) and noticed that the only result which is 'out of range' is the AST, the AST may actually be of cardiac origin and not from the liver at all, even though it has been reported under the liver test subheading.

Note: We have established that some blood tests presented as 'out of range' are in fact false positives; and some results are presented as 'in range' when they shouldn't be (false negatives). You should also bear in mind that we often get blood results which are 'out of range' under a particular subheading even though they don't belong to that group (as seen in the AST liver example).

Input and output

It may be helpful to consider organs and systems as having an 'input' or 'in' and a subsequent response of 'output' or 'out'. This can also help to discern family groups and can be presented either as a table or as a network diagram.

Table 3.1: Examples of input and output linking systems

Input	System	Output
Oxygen/Carbon Dioxide	Kidney	Erythropoietin (EPO)
Erythropoietin (EPO)	Bone marrow	Red blood cells
Cellular damage	Liver	C-reactive protein (CRP)
C-reactive protein (CRP)	Bone marrow	Neutrophils (White blood cells)
Unconjugated bilirubin	Liver	Conjugated bilirubin
Antigen	T-cell lymphocyte	'Wanted Poster' complex
'Wanted Poster' complex	B-cell lymphocyte	Antibody
Thyroid-stimulating hormone	Thyroid	Thyroxine

Draw a network diagram to show oxygen levels and kidney production of EPO, which induces the bone marrow to make more red blood cells which should carry more oxygen around, thereby reducing the need for more EPO. You can annotate this by adding a family group onto each organ or system.

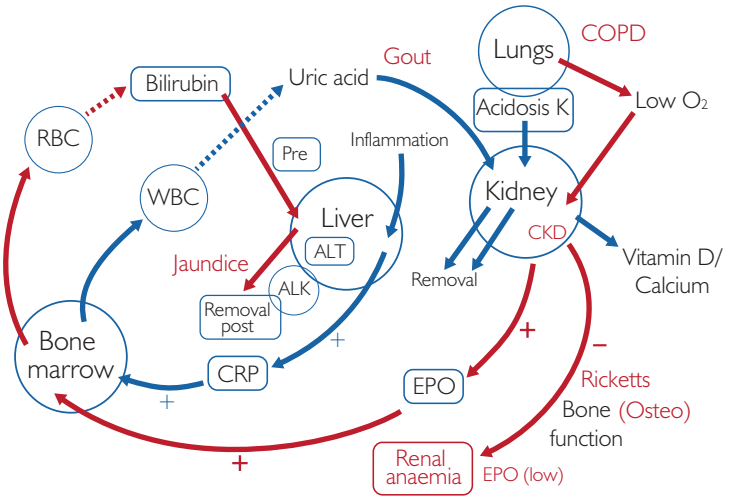


Figure 3.1. An example network diagram

This allows you to visualise different scenarios. What if the kidney is dysfunctional, for example? What if the patient has COPD or a bone condition?

Self-assessment

Draw your own network diagram, outlining inputs and outputs for your setting and annotating with relevant family groups.

4

Case study: Interpreting abnormal results

There are numerous, comprehensive case study textbooks available. However, even these may not be relevant to a complex, chronic patient or caseload, as is often found in primary care. Therefore, instead of placing numerous, partly relevant case studies throughout the text, one sample case study is presented here. Based on a real patient's test results, it will help you think about general strategies when interpreting a complex, chronic patient. These strategies can be applied in any clinical setting.

The following case study will enable you to:

- consider reference ranges
- look at family indices
- explain why bloods are selected for patient management.

This is a female patient, aged 60, who is being treated for rheumatoid arthritis (RA) with the drug methotrexate. The HIGH/LOW values are just outside the reference range. The patient has been tested every four weeks for the last year. Test 1 and 2 (shown below) therefore took place four weeks apart. RA symptoms were present and the patient had no other symptoms.

Table 4.1: Case study test results

Test	Test 1 result	Test 2 result
Full blood count (FBC)		
Platelets	NORMAL	NORMAL
White cell markers		
White blood cell count (WBC)	HIGH	HIGH
Neutrophils	HIGH	HIGH
Lymphocytes	NORMAL	NORMAL
Monocytes	NORMAL	NORMAL

Basophils	NORMAL	NORMAL
Red cell markers		
Haematocrit (Hct)	LOW	LOW
Haemoglobin (Hb)	NORMAL	NORMAL
Red blood cell count (RBC)	NORMAL	NORMAL
Mean cell volume (MCV)	NORMAL	NORMAL
Blast cells	NORMAL	NORMAL
Inflammatory markers		
Plasma viscosity (PV)	HIGH	NORMAL
Erythrocyte sedimentation rate (ESR)	HIGH	NORMAL
C-reactive protein	HIGH	NORMAL
Urea and electrolytes (U&Es), Kidney function:		
Sodium	NORMAL	NORMAL
Potassium	NORMAL	NORMAL
Urea	NORMAL	NORMAL
Creatinine	NORMAL	NORMAL
eGFR	NORMAL	NORMAL
Liver function tests (LFTs)		
ALT	NORMAL	NORMAL
Alk Phos	NORMAL	NORMAL
Bilirubin	NORMAL	NORMAL
Albumin	NORMAL	NORMAL
Globulin	HIGH	NORMAL

Interpreting the case study results

Some of the blood test results for the patient in the case study are out of range. We will work through the following questions in order to interpret these results:

- What values are abnormal for the condition?
- What would the blood tests from a typical RA patient look like?
- Why have these blood tests been requested?

Finally, we will apply the strategy.

What values are abnormal or typical for the condition?

An initial observation is that some conditions (like RA) can be cyclical, and the blood tests may not always match the clinical symptoms (see Test 2). Yet these false negatives could be problematic if this was a one-off blood test.

Looking at the values that are abnormal, we can make the following observations.

The white cell count is high. This count is raised in response to inflammation caused by an autoimmune response, some blood cancers and infection. The values observed, the duration of the condition and the presenting symptoms rule out a bacterial or viral infection. Given that red blood cell count (RBC) and platelet production is normal, a bone marrow dysfunction (leukaemia or myeloma) is unlikely. Hence these values are consistent with RA.

The neutrophils are high. Neutrophil numbers increase in response to sustained production of chemoattractants (which attract cells) like C-reactive protein (CRP) and TNF-alpha. CRP is raised relative to inflammation. Inflammation is cellular damage. Therefore CRP will rise in an autoimmune response through to severe burns and trauma. In practice, neutrophil increase is associated with bacterial infection, which causes inflammation and an increase in CRP. Neutrophils directly destroy the bacteria in the blood. In the case study, CRP and neutrophil values, the timeframe of the tests (one year) and presenting symptoms make a bacterial infection unlikely. The values are therefore consistent with an autoimmune response.

The haematocrit (percentage of red blood cells in the whole blood) is low. However, there are no symptoms of anaemia, the value is very close to the reference range 0.353 (low range 0.360) and crucially it doesn't fit with the other markers of red cell status. RBC and haemoglobin (Hb) are both normal. The value is not changing over time. A false positive is likely, and the result could be described as 'not clinically significant at this time'.

Plasma viscosity (PV) is high. PV is a marker of 'extra stuff' in the blood. In this case, it is raised due to increased white cells (neutrophils) and globulins (antibodies).

Globulins are raised. Globulins are a crude, surrogate measure of antibody production. The globulin count is raised when antibodies are produced, and antibodies are produced by B cells (a type of lymphocyte). Globulin increases in viral infections (a normal response by the lymphocytes to infection). But the case study patient's lymphocytes are normal and she

has no symptoms. The globulins are raised in a myeloma (dysfunctional B cells), but again the lymphocyte count and globulin value make a diagnosis of myeloma unlikely. (A Bence Jones protein or electrophoresis band will confirm.) Globulins are often raised in an autoimmune response. Because the body makes antibodies to 'itself', this will not usually increase the lymphocyte count (as would be seen if the antibodies were being produced against a large viral infection, driving up lymphocyte number and antibody production).

Why have these blood tests been requested?

This woman is typical of the patients encountered in practice – she has several different conditions, for which she is taking several types of medication. Looking at the long list of tests can be overwhelming and confusing. A useful strategy is to split the blood tests into a 'hierarchy of conditions' and discuss which ones require red flags (to be actioned quickly), referrals, interventions and repeat tests.

To monitor RA, the white cell count, neutrophils, PV, CRP and globulins have been requested. Any significant change in these values could indicate an additional bacterial infection. Thus, this suite of blood tests may be monitored by the secondary care specialist consultant for RA and by the primary care practitioner for additional infections.

To monitor the effects of methotrexate, two suites have been requested: LFTs, U&Es and FBC (red cell markers). Methotrexate can be toxic to some patients and the LFT panel will provide a red flag if this is the case. This also explains why the patient has a blood test every four weeks.

Methotrexate works in autoimmune conditions by restricting folate availability in the blood. Folate (folic acid) is the 'currency' the cells use to replicate (multiply) and grow (hence methotrexate's use as an anti-cancer therapy). Long-term restriction of folate can lead to macrocytic (high red cell volume) anaemia because red blood cell lifespan is only 12 weeks. To monitor this long-term predisposition to anaemia, RBC, Hct, Hb and MCV are measured regularly.

As a differential diagnosis, platelets (and RBC) are measured for possible bone marrow dysfunction, which could be masked by increased white cells in the autoimmune response.

U&Es are measured as a general marker of kidney function; and potassium is a red flag for a cardiac event. As a differential for anaemia, is any possible anaemia linked to renal insufficiency? Finally, U&Es are measured (in the case study) as a differential diagnosis for the LFTs. Some liver enzymes, like alkaline phosphatase (Alk Phos or ALP), are also found in the kidney. A raised ALP and normal U&Es can quickly indicate that the liver is the likely origin and cause for investigation.

Applying the strategy

Let's think about family groups and the four questions to apply to these results.

The initial family group in the FBC, and a great place to start with any blood test, is Hb for red blood cells, then white cells, then platelets.

- Look at Hb – this is in range.
- Now look at Hb's two mates (red cell indices), HCT and RBC. The only one out of range is HCT.

Now let's apply the four questions to HCT:

- How far out of range is it? Not very. It's borderline. In fact it's been borderline for some time and is often in range normal.
- Is the patient symptomatic? No.
- Is this an important blood test? No, the HCT result doesn't usually change the management of the patient.
- What do its mates say? Well, RBC and Hb are both in range so they don't support HCT.
- We will therefore ignore HCT as a false positive.

Next, check platelets:

- They are in range so the bone marrow is likely to be working OK.

Next, look at the WBC:

- The WBC is out of range – elevated.
- What type of WBC is causing this increase? Neutrophils.
- What switches on neutrophils? CRP.
- CRP is also out of range in a group (CDL), which is commensurate with autoimmune.

Now look at the four questions for the white cells:

- Are they far out of range? Yes.
- Does the patient have symptoms? Yes.
- Family group? Yes (WBC, neutrophils, CRP).
- Is this an important result? Yes.

Finally, mop up any additional bloods, such as globulin:

- Globulin measures antibodies so this could be due to a viral infection.
- But the lymphocytes (the WBC that fight viral infection) are not raised.

- The CRP isn't high enough, and the patient doesn't really have the expected symptoms.
- So the rise in globulins is much more likely to be caused by the presence of auto-antibodies, as seen in autoimmune (which this patient has).
- We only measured liver function because the patient is on a drug called methotrexate which can be toxic to the liver.
- So we now note the globulin result, which is actually nothing to do with the liver (as discussed).

To summarise, all the bloods (apart from HCT) are fairly 'normal' for a patient with rheumatoid arthritis (RA) and we can ignore the HCT as a false positive.

Self-assessment

Work through some case studies in your own setting and apply the four questions, family groups, input and output, and think about the four ways in which blood can change.

5

The full blood count

The full blood count (FBC) test provides information about the cellular components of the blood. The FBC can be used in detecting anaemia, understanding inflammation and infection, and monitoring coagulation and leukaemia presentation.

The FBC result is usually made up of three components: the red cell indices, the white cell indices and the platelets. Table 5.1 (below) shows how red blood cell count (RBC), white blood cell count (WBC) and platelets alone can initially be used to stratify some indicative conditions (matched to symptoms).

Table 5.1: Full blood count sample results

Condition	RBC	WBC	Platelets
Anaemia	LOW	NORMAL	NORMAL
Infection	NORMAL	HIGH	NORMAL
Autoimmune conditions	NORMAL	HIGH	NORMAL
Leukaemia	LOW	HIGH	LOW
RA, methotrexate, no folate	LOW	HIGH	NORMAL
Aplastic anaemia, bone marrow failure	LOW	LOW	LOW

The red blood cell indices

These will usually be haemoglobin (Hb), haematocrit (Hct) and RBC.

Hb is the oxygen-carrying protein in red blood cells (erythrocytes) and it can be used as a marker for surgical outcome. It can also be compared to other red cell indices, such as anaemia (Hb low). In addition, it can be used as a surrogate marker of renal insufficiency (low), blood loss (low), oxygen deprivation (high) or glucose in excess (HbA1C), and in genotyping for some sickle cell conditions.

The red blood cells transport oxygen, carbon dioxide and some nutrients between organs, and they have a lifespan of around 12 weeks. This is important when thinking about *repeating* or measuring an intervention. In a patient who is being treated for iron deficiency anaemia, there may therefore be little value in repeating the RBC before 12 weeks. If the patient is being corrected for a surgical intervention, because their Hb is too low or following blood loss, then cells called reticulocytes can be measured. Reticulocytes are 'baby' red blood cells and their profile increases to intervention for anaemia within a few days (compared to a few weeks for mature red blood cells).

Production of red blood cells is regulated by a biochemical process called erythropoiesis. This process is linked to tissue hypoxia (lack of oxygen) and driven by the hormone erythropoietin (EPO), which is produced by the kidney (◀▶ U&Es and renal anaemia). An elevated Hb and RBC are called polycythaemia. This is often caused by tissue hypoxia due to carbon monoxide or carbon dioxide, cigarette smoking, asthma or chronic obstructive pulmonary disease (COPD) or being at high altitude. Alternatively, high red cells may be seen in damage to the spleen (caused by trauma) or acute dehydration (relative rises). In practice, this normal physiological process is competing with disease pathologies and conditions. These are shown in the figure below, with the orange arrow representing COPD and the green arrow representing renal dysfunction.

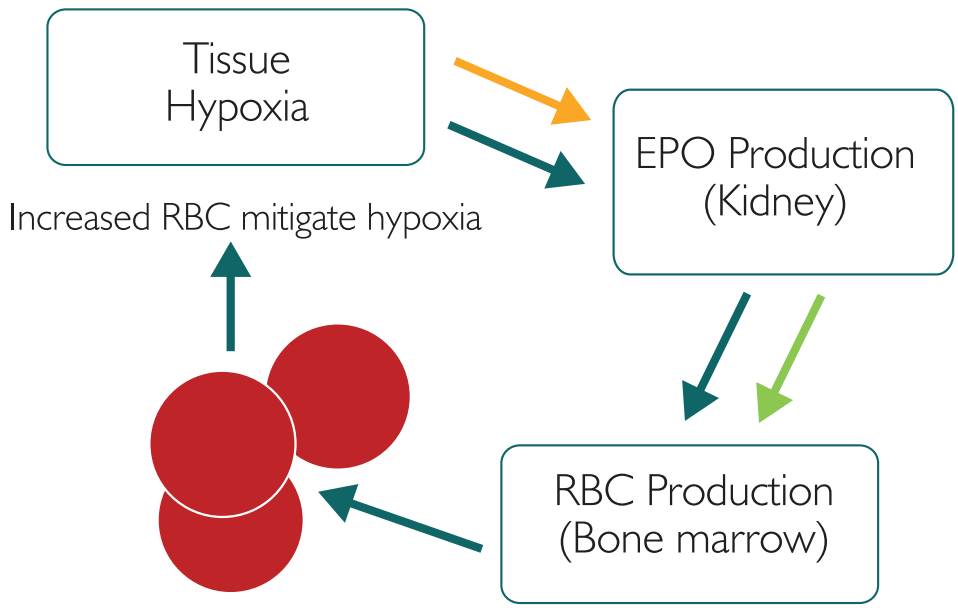


Figure 5.1: The process of erythropoiesis, with COPD and renal dysfunction

Considering the orange arrow, COPD causes narrowing of the airways and a subsequent fall in airflow. Bronchitis or emphysema (often caused by exposure to particulates such as cigarette smoke) is usually irreversible and leads to a lack of oxygen. This will usually cause hypoxia, which will result in increased EPO. EPO induces bone marrow production of red blood cells (erythropoiesis) and thus an increase in RBC. However, given the chronic, ongoing nature of COPD, this rise in RBC will not usually mitigate the hypoxia. COPD patients will often therefore have higher than normal RBC, which may present an increased risk of a cardiovascular event. Oxygen therapy may help to reduce or suppress the red blood cell production.

Considering the green arrow, overall renal function, as measured by estimated glomerular filtration rate (eGFR), generally falls by 10% per decade after the age of 35. Given this natural decline in renal performance, the kidney's ability to sustain production of EPO is also reduced, leading to the renal anaemia seen in some elderly patients. EPO supplements may help to induce red blood cell production in these cases.

Anaemia

Anaemia means a reduced RBC (and Hb) and could indicate inability to sustain EPO (◀▶U&Es), bone marrow insufficiency (leukaemia – ◀▶ white cell and platelets as described, see Table 3.1 above), blood loss or nutritional anaemia.

Low Hb/RBC/lethargy could be anaemia, especially with normal white cell and platelet number, but what type of anaemia? Mean cell volume (MCV) can help identify the type of anaemia. MCV represents the average size of the red blood cells – big, small or normal.

Normal MCV (normocytic) means that the body can make red cells but it cannot keep them. This could suggest blood loss (due to menstruation, surgery or trauma, for example) but it can also be a surrogate marker of stomach ulcers or GI cancers.

Low MCV (microcytic) means that the body does not have the structural components needed to make red cells of the correct size. **Storytelling: Imagine not having enough metal tent poles for your tent. The tent will not be the right size and will be small.** This is usually due to an iron (Fe) deficiency, so consider tests for serum ferritin and total iron-binding capacity (TIBC). Possible causes of this could be Fe deficiency, sickle cell or thalassaemia.

High MCV (macrocytic) means that the body does not have enough nutritional components to make red cells of the correct size. Consider tests for either folate or B_{12} deficiency. Folate and B_{12} in this pathway provide 'instructions' for the cells by making accurate DNA. **Storytelling: Imagine owning a new inflatable tent, which requires 75 litres of**

air to make it the right size. If the instructions have a misprint of 95 litres then more air is added and the tent will be bigger than expected. Possible causes of this could be liver disease, pregnancy, some antibiotics, and folate and B_{12} deficiency.

If folate is low, it could be due to dietary factors or a genetic defect in the enzyme (MTHFR) or some medications such as anti-cancer or epileptic drugs, or alcohol intake (◀▶ deranged LFTs). Alcohol can affect the stomach lining and reduce folate absorption. As red blood cells last for around 12 weeks, it would take sustained alcohol intake to change the entire RBC profile.

If B_{12} is low, it could be dietary or could be due to an autoimmune pernicious anaemia (PA), which can affect the stomach lining and reduce oral B_{12} absorption. A positive parietal cell and/or intrinsic factor antibody test can confirm PA. Given that oral absorption is restricted in PA, an intra-muscular B_{12} injection is usually required. Failure to correct B_{12} deficiency may lead to B_{12} neuropathy and demyelination because B_{12} is required to make the myelin sheath. If the myelin sheath is dysfunctional then nerve integrity is lost and neural and vascular retraction from the site (usually feet) will occur. ◀▶ Patients who are positive for parietal cell or intrinsic factor antibodies may also be positive for anti-thyroid antibodies and have low T4 (thyroxine). A thyroid function test (TFT) may therefore be appropriate.

In summary, using the MCV test from the FBC can help to differentiate between microcytic, normocytic and macrocytic anaemia.

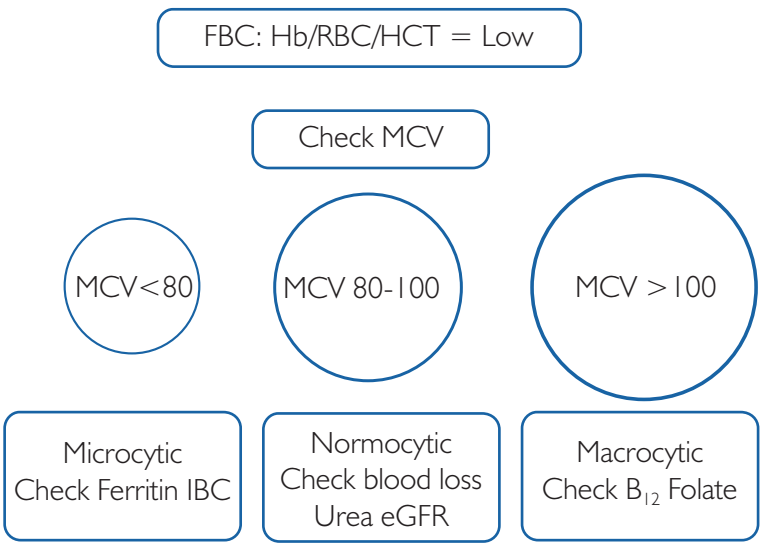


Figure 5.2: Patterns of Anaemia

Anaemia of chronic disease

Some patients with long-term chronic conditions, particularly ones with an underlying inflammatory disorder such as COPD, may develop anaemia of chronic disease (ACD). In the normal red cell model, iron (Fe) is stored as ferritin. In states of low ferritin, the capacity to bind iron increases. This is called total iron binding capacity (TIBC). In a normal dietary or gastrointestinal iron-deficient anaemia, the patient will usually have a low MCV and low ferritin, but a high TIBC. In ACD, the route by which the stored iron is utilised to make red cells is compromised or broken. The cell has plenty of stored iron so it doesn't need to bind any more. Thus, in ACD serum ferritin is often high and TIBC is low. But the iron is locked in, so the cell can't use it to make red blood cells.

We need ferritin, EPO (from the kidney), B_{12} and folate to make red blood cells. If any of these ingredients is lacking, the patient will make fewer red blood cells (in other words, they will develop anaemia). Each of these missing ingredients will have a particular effect on the red blood cells. For instance, a lack of iron will mean fewer red cells and the cells will also be smaller. A lack of B_{12} will mean fewer red cells and they will also be bigger.

To summarise, we can use MCV and we can consider the size of the red blood cells and which additional blood tests to request. Don't forget about the overhang, which was mentioned earlier. An MCV of 99fL, which is close to the cut-off of 100 could really be a macrocytic anaemia. (Measuring B_{12} and folate could confirm this.) Finally, note that MCV is a mean – so if a patient has a mixture of sizes, this can affect the mean. Measuring the iron, B_{12}, folate, etc. can help to confirm which type of anaemia the patient has.

Neuropathies

In addition to pernicious anaemia, B_{12} neuropathy (low RBC, low B_{12}, high MCV, normal LFT) and diabetic and alcoholic neuropathies may co-compete in practice. Each has a different pathology.

In diabetic neuropathy (high HBA1C, normal RBC), glucose in excess binds directly to the nerve and blocks nerve impulses.

Alcoholic neuropathy (low RBC, high MCV, low folate and high LFTs) is due partly to direct poisoning of the nerve by alcohol, and by lack of nutrients caused by alcoholism (B_{12} and proteins for the myelin sheath and folate). Folate has a number of roles in the body. We have discussed DNA creation, but it is also linked to clearing an amino acid called homocysteine (HCY). High levels of HCY are thought to affect vascular tone, making blood vessels stiff and reducing blood flow. This explains why poor diet and excessive alcohol intake are linked to peripheral vascular disease. Given its effect on vascular tone, high HCY is also a factor in gout, poor healing of fractures and infections, and cardiovascular disease.

Finally, the haematocrit (Hct) is often used as a confirmatory marker of RBC status. However, Hct is a crude marker, as it represents the proportion of the whole blood made up of red blood cells. This is usually 35–55% (or 0.35–0.55) and has been used historically because it is very cheap and very quick. Hct goes down in anaemia, and up in polycythaemia.

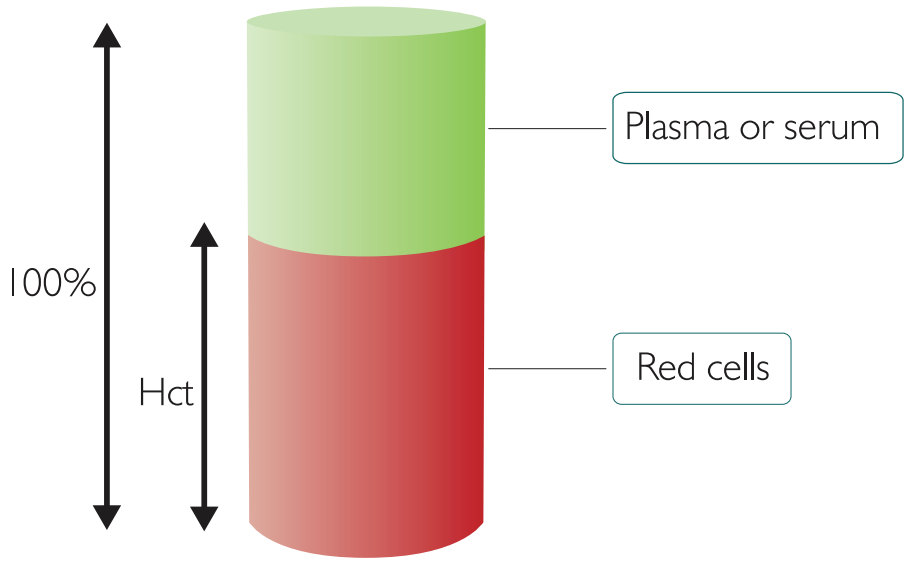


Figure 5.3: Haematocrit as a proportion of whole blood

Dementia

Given that anaemia can produce some of the symptoms of dementia, it may be appropriate to screen for this, looking for low RBC or low Hb. Other exclusions include: raised inflammatory markers (erythrocyte sedimentation rate, C-reactive protein and plasma viscosity) which may indicate pulmonary vasculitis, especially with a positive ANCA test (anti-neutrophil cytoplasmic antibodies); hypothyroidism via a thyroid function test; glucose control and diabetes; and a human immunodeficiency virus (HIV) or syphilis screen.

The white blood cell indices

Many aspects of white blood cells have already been discussed in the case study in Chapter 4. Further clinical practice examples will be explored in Chapter 7 (inflammatory markers), Chapter 8 (autoimmune conditions) and Chapters 10, 11 and 12 (chronic disease markers).

White blood cells (leukocytes) respond to, prevent and remove infection, and they are central to the inflammatory response via C-reactive protein (CRP) and TNF-alpha. There are different types of white cells, which can help identify the pathology. The white cells can either be presented as a count (a number) or as a percentage – say, 80% of the white cells are neutrophils. The percentage of white cells is called the differential or WCD.

Low WBC is a fairly rare condition (leukopenia), which is sometimes seen in patients undergoing chemotherapy. It is also found in aplastic anaemia (this is total bone marrow failure so patients will also have low RBC and low platelets), HIV (especially the T cell lymphocytes), some cases of lupus, and copper and zinc deficiency.

High WBC has three common causes, although the presenting symptoms will be different: leukaemia (usually with low RBC and low PLT), infection (look at type of white cell) and autoimmune conditions (usually raised neutrophils). ◀▶ Inflammation markers such as erythrocyte sedimentation rate (ESR), PV and CRP.

The main role of neutrophils (WCD 40–80%) is protection from bacteria and they are driven (increased) by markers of cellular damage such as CRP. They are therefore often used in autoimmune, inflammatory monitoring.

The main role of lymphocytes (10–20%) is protection from viral infection. There are two types: B cells (antibody production) and T cells (viral identification and destruction). T cells also play a role in misidentifying self-tissue in autoimmune conditions.

Monocytes (5%) infiltrate tissue in systemic bacterial tissue. Once in the tissue, they are called macrophages, and they can also incorporate low-density lipoprotein (LDL) and become a foam cell. Foam cells can then form an atherosclerotic plaque and increase cardiovascular disease risk (◀▶ cholesterol and TG concentrations and chronic disease management).

Eosinophils and basophils (5%) ensure protection from some parasites and may form a response in asthma, allergy and hypersensitivity conditions such as irritable bowel syndrome (IBS).

Blast cells (atypical, < 2%) can indicate bone marrow dysfunction. These are white cells that cannot be identified because they have been incorrectly made, and so they are often seen in leukaemia, myeloma and lymphoma.

A lymphoma is another cancer of the B cell and/or T cell lymphocyte activity, but damaged cells usually aggregate in the lymph nodes. Thus, patients are often immune-compromised, with reduced antibody production. The next step is to determine the type of lymphoma. Hodgkin's is a positive for a subset of the B cell called a Reed-Sternberg cell, which may require different treatments from non-Hodgkin's. Lymphoma patients usually have recurrent infections and are negative to Bence Jones paraprotein.

6

Coagulation and deep vein thrombosis

Coagulation markers are usually used pre-operatively to prepare patients for surgery, to manage coagulation medications and to assess venous thromboembolism (VTE) risk. The usual panel will be platelets, fibrinogen, prothrombin (PT), activated partial thromboplastin time (aPTT) and international normalised ratio (INR), and D-dimer.

Venous thromboembolism

A VTE is a clot (thrombus) in the vein, usually a deep vein thrombosis (DVT), which is dislodged (an embolism) and can move to heart, brain or lungs (pulmonary embolism or PE), with serious consequences.

VTE is most commonly caused by poor post-surgical management, or by poor lifestyle (such as a high-fat diet and sedentary habits). A VTE or DVT is diagnosed by means of a history, physical manipulation, scoring and blood tests. A combination of the ultrasound scan (US), Well's score, Homan's test and a D-dimer blood test will usually indicate a DVT diagnosis. In the UK, NICE guidelines and local operating procedures should be followed for VTE diagnosis.

The Well's score (WS) is a history-taking scoring system that takes into account active cancer, whether the patient is bedridden or has had major surgery, calf swelling in one leg, leg swollen, tender deep vein/groin, and previous DVT. The higher the Well's score, the more likely it is that a DVT may be present.

D-dimers (Dd) are fragments of a clot, which has broken down under the action of plasminogen. They are therefore indicative of a large unstable clot being resolved (DVT). This test is often used in conjunction with the Well's score[1]:

- High WS, then Dd less relevant and treatment initiated
- Medium/Low WS, negative Dd, then unlikely DVT
- Medium/Low WS, positive Dd, then usually ultrasound scan.

[1]These statements are examples of context and clinical advice to explain DVT, Well's score, ultrasound and D-dimer blood test. For patient management in the UK, see NICE VTE guidance.

The D-dimer test can also be used following treatment, to confirm that the clot has been fully resolved. As VTE and DVT treatment is fairly well tolerated by most patients (and in view of the complications that may result from misdiagnosis or delay), the treatment is often given on symptom presentation alone.[2] Current guidelines propose an ultrasound within four hours.

Raised platelets (thrombocythaemia) can also lead to increased risk of VTE and DVT. Raised platelets can be caused by myeloproliferative diseases such as chronic myeloid leukaemia (CML) and polycythaemia vera. These cause over-production of platelets by the bone marrow. A positive Philadelphia chromosome confirms CML.

Coagulation monitoring

Coagulation is a clotting process that is usually initiated by tissue damage and augmented by the platelets. It has two components: intrinsic (driven by platelets) and extrinsic (driven by tissue). Both ultimately lead to fibrinogen and then to fibrin, which forms a clot complex with the platelets. (◀▶This process also produces an inflammatory response, so it makes sense also to measure white blood cell count.) This leads to an increase in erythrocyte sedimentation rate (ESR) through increased fibrinogen-sticking red blood cells.

Warfarin generally works by suppressing the liver's production of factors that induce clotting. This process is extrinsic, as it is in the liver and outside the blood. It is also relatively slow, as it takes up to five days for warfarin to work.

The usual blood test for warfarin is the international normalised ratio (INR), which essentially measures how long it takes the blood to clot in a lab, converted to an arbitrary value of 1. If, following warfarin, the INR rises to 2, the blood is therefore taking twice as long to clot. Generally, an INR greater than 4 is problematic for bleed management, and may affect decisions about micro-surgery and injection interventions.

The INR is ultimately a marker of fibrinogen production, and thus liver function (as this is where fibrinogen is made). It is also used as a surrogate marker of liver function in patients who are not on warfarin. If INR is measured in an alcoholic patient (it is quick and cheap) and the result is 7, liver dysfunction would be highly likely.

Heparin, clexane, deltaparin and low molecular weight heparin work via an intrinsic pathway, inside the blood, by directly binding to (and thus deactivating) chemicals that induce a clot. These medications will therefore work very quickly (within a few minutes) and can also be used as a prophylaxis. In view of their quick action, they are often given as a dual therapy

[2]These statements are examples of context and clinical advice to explain DVT, Well's score, ultrasound and D-dimer blood test. For patient management in the UK, see NICE VTE guidance.

with warfarin, as the latter takes a few days to work. In primary care, blood tests are less likely to be ordered for patients on these types of therapies. The test to assess these interventions is called aPTT and is usually ordered before invasive intervention.

The final group of therapies work on the platelets so they are intrinsic and can be assessed by aPTT. These include aspirin and clopidogrel. The drugs are converted in the liver into pro-drugs, which remain in production for a few days after the oral base drug has ceased. The platelets express sticky surface proteins (like a Velcro coat), which enable them to stick together and clot. This protein is called Von Willebrand's factor (VWF) and it is suppressed by the pro-drug. Once suppressed, it is likely that the platelets will have suppressed clotting ability for their lifespan of up to two weeks. Patients on high doses of such treatments may need platelet replacement to prevent bleeds – though not while the pro-drug is being made by the liver, as this will simply affect the newly transfused platelets.

Alcohol works on both intrinsic and extrinsic pathways. ESR can also be used to indicate cancer, myeloma and macrocytic anaemia and so could be being used for this reason. ESR also increases with age. **Storytelling: Acutely, alcohol 'gets the platelets drunk' by suppressing their VWF activity. This makes them take off their sticky coats and reduces their ability to stick together. Hence, sword fighting after drinking wine is not a good idea!** In alcoholic patients, this acute phase is augmented by liver dysfunction – the liver does not produce fibrinogen and clotting times increase significantly.

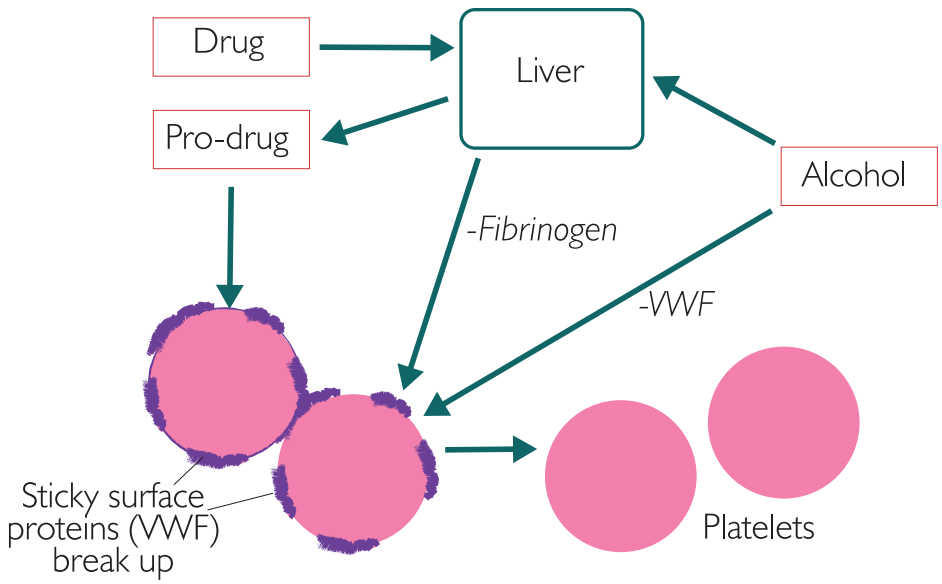


Figure 6.1: The interaction between alcohol, drugs and the liver

Finally, certain haemophilic conditions (such as an absence of VWF or of factors such as Leiden V) will also increase clotting time. In the UK, most inborn errors of metabolism and coagulation are seen in neonates. Patients with sickle cell anaemia or thalassaemia may also have a shorter red cell lifespan. This may give them a predisposition towards abnormally shaped cells, which are likely to stick together, forming a clot. The clot may then cause a painful 'crisis' and in some cases a stroke.

7

Inflammatory markers

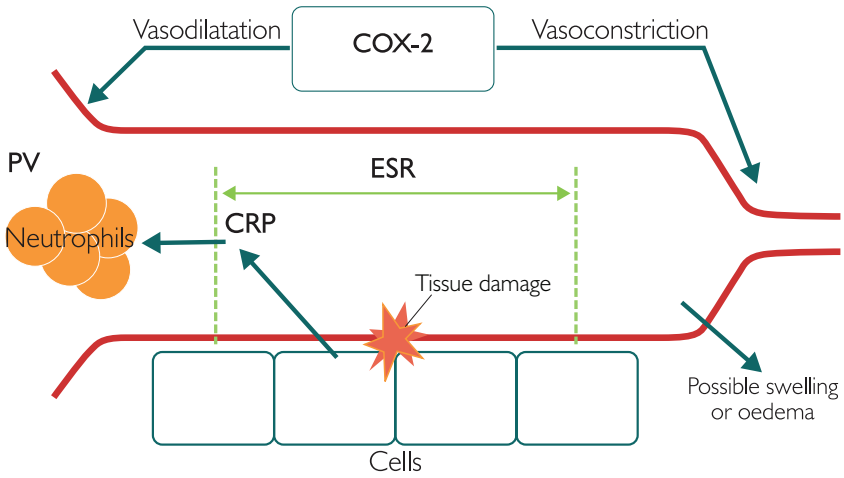
A typical inflammatory marker panel would be erythrocyte sedimentation rate (ESR), C-reactive protein (CRP) and plasma viscosity (PV). Each test tells us something different about the inflammation pathway, and some tests are more appropriate than others.

There are numerous inflammatory markers. In practice, these tests tend to be requested in two ways: either as a typical inflammatory marker panel (CRP, ESR, PV); or as a specific test, given the new interest in this area for research and drug treatments. Inflammatory markers are usually cellular signalling molecules (signal cells) or adhesion molecules (which make cells stick to the vascular wall). These adhesion molecules include tumour necrosis factor alpha (TNF alpha), interleukin-6 (IL-6), iCAM and vCAM, which are the basis of advanced drug treatments.

Inflammation is a response to cellular damage and the inflammatory markers generally rise in relation to the degree of tissue damage, particularly CRP (see the Chapter 4 case study). Cellular damage can be caused by infection (bacterial and/or viral), an autoimmune response, cancer and trauma.

An initial strategy is to map the inflammatory markers to interventions and symptoms. For example, if investigating a possible infection a few days after a hip or knee replacement, consideration should be given to the significantly raised inflammatory markers resulting from the surgery. A blood culture may be more appropriate. Although in practice an antibiotic is often prescribed, this may not be needed.

In inflammation resulting from cellular damage, there are two phases – cellular and exudative. Cellular inflammation is controlled by CRP and TNF-alpha and is the cellular response. Exudative inflammation is controlled by an enzyme called cyclo-oxygenase 2 (COX-2). This enzyme makes prostaglandins that modulate vasoconstriction and vasodilatation (the swelling response). COX-2 can be partly inhibited by aspirin and non-steroidal anti-inflammatory drugs (NSAIDS).



ESR – erythrocyte sedimentation rate PV – plasma viscosity
CRP – C-reactive protein

Figure 7.1: Cellular and exudative inflammation

The tissue damage elicits a cellular signal: CRP, to recruit and increase the number of neutrophils. To aid this process, the enzyme COX-2 produces prostaglandins, which cause constriction and dilatation at the site to affect blood flow. This response may cause pain (if a nerve is part of the constricted area), swelling, oedema and redness of skin.

Table 7.1: Inflammation analogies

Test: C-reactive protein (CRP)

Storytelling:
CRP is the 'fire alarm'.
The more damage there is, the more alarms will sound and the more people will ring the fire brigade.
Once the fire-fighters are on site, the alarms may be switched off.

Interpretation:
CRP is relative to the degree of cellular damage. The more invasive the bacteria or trauma, the more CRP is produced.
If the blood test was requested post-injury, the CRP signal may be lost because the site of injury has been repaired.

Test: Neutrophils

Storytelling:
Neutrophils are the 'fire engines' that respond to CRP and cellular damage. The more damage there is, the more fire engines will be called.
A fire engine can also attend a fire without being called out, perhaps because the crew members have spotted a small fire on waste ground on the way back from a call (bacteria).

Interpretation:
Neutrophils respond to cellular damage but can also spontaneously destroy bacteria.

Test: Plasma viscosity (PV)

Storytelling:
As the fire engines and debris build up in the surrounding streets, the pressure in the area will increase. The more fire engines (neutrophils) and news reporters (bacteria, for example) there are, the more blocked the street will become.
Even after the fire alarm (CRP) has been switched off, the engines may stay at the site for a while. Neutrophils have a lifespan of two weeks so PV may be elevated after the initial CRP value.

Interpretation:
PV is a crude marker of material in the blood that increases pressure and 'thickness' or viscosity. The more cells and bacteria there are, the higher the PV.
PV can be useful to show that tissue injury occurred in the absence of a raised CRP.

Test: Erythrocyte sedimentation rate (ESR)

Storytelling:
Following a large fire, structural reinforcement is required. ESR indicates the amount of clotting around the damage site.

Interpretation:
In some autoimmune conditions that have relatively small amounts of tissue damage, such as rheumatoid arthritis (RA), ESR may be requested on a six-monthly basis because the damage is too small to affect the ESR.

Test: White blood cell count (WBC)

Storytelling:
The white cells are the 'emergency vehicles' (police cars and fire engines). As more fire engines (neutrophils) take to the street, the number of emergency vehicles on the roads increases. The WBC is the total number of white cells in circulation.

Interpretation:
WBC reflects inflammation. ◀▶ CRP, PV, ESR and white cell type. Increased neutrophils are likely to indicate a bacterial infection or an autoimmune response. Raised lymphocytes are likely to be due to viral infection.

Erythrocyte sedimentation rate

The erythrocyte sedimentation rate (ESR) is 'how far red cells (erythrocytes) fall (sediment) in a tube, in an hour'. As the tissue is damaged, fibrinogen is released. Fibrinogen 'sticks' red cells together, making them heavy, so they fall further in the test. A high ESR means that more red cells are stuck together by fibrinogen, which results in more cell/tissue damage. ESR is usually raised in inflammation, but it can take time for this 'sticking process' to occur and indeed be removed. ESR is therefore not as quick to respond to damage as CRP. In some cases, ESR will rise in anaemia (independently of fibrinogen and inflammation). Large, macrocytic red blood cells will fall more quickly than small or normal-size ones. ESR can also be used to indicate cancer, myeloma and macrocytic anaemia and so could be being used for this reason. ESR also increases with age.

Storytelling: If you went to the top of a tall building and dropped three footballs (red cells), timing how long they took to fall, that would be a normal ESR. If you repeated this exercise, but placed the footballs in a heavy sack (fibrinogen resulting from inflammation), the balls would fall faster. However, as discussed earlier, ESR can also be independent of inflammation. If you repeated the demonstration, but dropped a large medicine ball (a macrocytic red blood cell) instead of three footballs, the large ball would fall faster. In other words, ESR can also be raised in macrocytic anaemia. You could cross-check ESR with mean cell volume (MCV) to find out the size of the red cells.

Plasma viscosity

Plasma viscosity (PV) is a measure of pressure *in* the blood (not blood pressure). It is measured in mPascals, a unit of pressure. PV is a surrogate marker of material that is not expected to be in the blood. As more white cells, bacteria, antibodies, red cells, in fact any material, build up in the blood, the pressure will rise and so will PV. It's often described as 'thick blood' because more material is present.

Storytelling: Visualise a glass filled with water, and imagine stirring it with a spoon. The amount of pressure or power needed to stir the water is a normal PV. If you add a handful of marbles to the glass, it will be harder to stir because more pressure will be needed. Now imagine the glass is inside a box so you can't see what is causing it to get harder to stir. It could be marbles (red blood cells), but it could also be stones (white blood cells) or woollen fibres (antibodies) or even sand (bacteria). Hence, an increased PV demonstrates an increase in pressure within the blood. But to determine the cause you'll need to cross-check with other tests such as full blood count (FBC), looking at WBC, RBC and so on.

PV is mainly used to understand inflammation. It's used in autoimmune conditions because it can indicate that additional antibodies and white cells are present, thickening or increasing pressure in the blood. PV can also be useful for monitoring against a baseline over time. However, it has limitations in that it is not specific to one disease and is therefore a crude global marker.

CRP

C-reactive protein (CRP) is a molecule that attracts and induces production of white blood cells (usually neutrophils), following inflammation. CRP is a marker for inflammation and infection and it can be used in autoimmune conditions because it can represent cellular signalling. CRP is also being used for chronic disease surveillance, as it is sensitive enough to represent vascular damage (raised CRP) and liver disease (low CRP). However, in some settings it can be expensive. Also, the signal can be lost as CRP is cleared, whilst ESR and PV may remain high for some time after the initial response.

The following storytelling offers a strategy to assist in interpreting a complex and dynamic process.

Storytelling: CRP is the fire alarm that goes off in the building, attracting fire engines (neutrophils/WBC) and causing a traffic jam in the street (PV). The structural support (fibrinogen scaffold) around the building is ESR. It's unlikely that a small fire, as in the specific connective tissue damage caused by rheumatoid arthritis, will result in the entire building needing scaffolding. ESR is therefore only requested periodically, usually every six months. However, following major surgery or a large muscle trauma, ESR would be required.

Anti-inflammatory treatments

Anti-inflammatory treatments work along the pathways shown in Figure 8.1 (see page 59). Anti-TNF alpha treatments suppress the initial chemoattractant; drugs like methotrexate work by suppressing the neutrophil recruitment and replication (it was originally an anti-cancer cell proliferation drug); and drugs like aspirin and NSAIDS work to inhibit COX-2 and relieve the vasoconstriction.

To summarise, ESR represents tissue repair post-injury (fibrinogen), PV reflects components in the blood due to inflammation (more WBC, more antibodies), and CRP represents the tissue signalling in response to cellular damage.

8

The immune system

In an autoimmune disease, our white cells destroy our own cells. The most common reason for this is incorrect cellular signalling, when the cell expresses a 'self-antigen'. Most autoimmune diagnostic tests are based on detecting the presence of self-antigen or auto-antibodies.

Normal response to viral infection

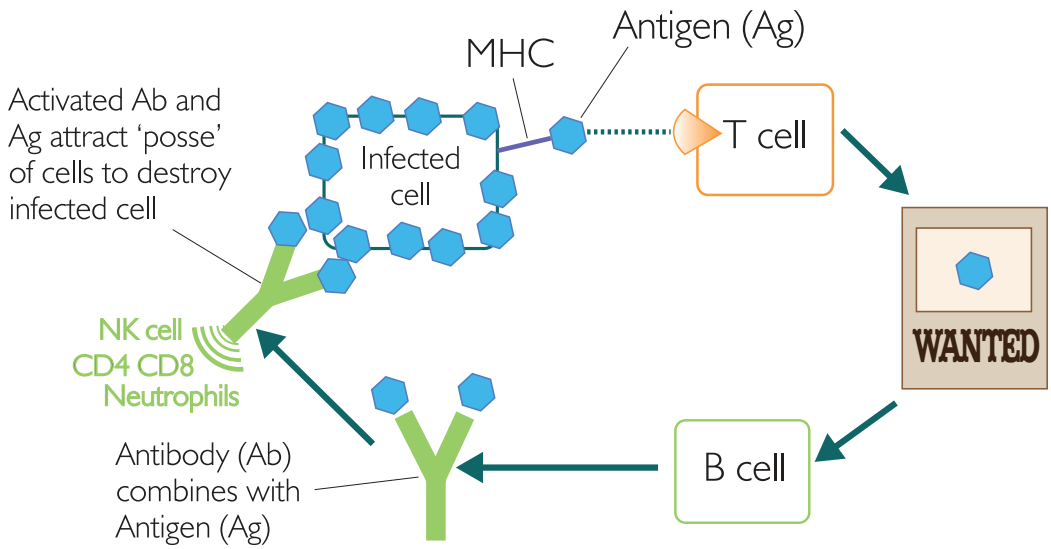


Figure 8.1: A normal response to a viral infection

In Figure 8.1, the virus uses the internal machinery of the cell to replicate itself. It then bursts the cell and infects an adjacent cell. Because the virus is small (compared to bacteria), the cell needs to make the white cells aware that it has been infected.

Storytelling: The virus inside the infected cell is too small to be detected by the white cells. The infected cell spreads the news of the infection to the white cells by taking a small part of the virus, called an antigen (Ag), and 'placing it

on a stick' outside the cell. This stick is called major histocompatibility complex (MHC). The T cell (a type of lymphocyte) then acts like a camera and takes a picture of the MHC/Ag. Having taken the photo, the T cell uses it to make a 'Wanted poster' of the MHC/Ag 'villain' and shows the poster to the B cell (a type of lymphocyte). The B cell makes antibodies (Ab) specifically designed to counteract the Ag, and the tailor-made Ab binds itself to the Ag. Once bound, a signal is activated and this attracts a 'posse' of neutrophils, natural killer cells and CD4/CD8 cytotoxic T cells to destroy the cell hosting the villainous virus.

The process of destroying the virus usually causes cellular damage. It increases C-reactive protein (CRP), produces more neutrophils and plasma viscosity (PV), and raises WBC (white cell or lymphocyte count). For example, human immunodeficiency virus (HIV) destroys the T cells and so the lymphocyte count may actually go down. With the T cell 'camera' not working, AIDS patients may be susceptible to otherwise benign viral infections.

This can also be seen when diagnosing viral hepatitis. For example, the presence of hepatitis B surface antigen (HepBsAg) may indicate a current infection. Likewise, the presence of HepB Ab will suggest a live infection (if HepBsAg is also present), a successful vaccination, or the presence of the disease in the past (as the B cells have made a specific Ab for HepB). There is also a new, rapid technique available that measures the DNA or RNA of the HepB virus directly in the cell. This is helpful in children or acute cases, as it may otherwise take a few days or even months for the patient to produce enough Ag or Ab to be measured (◀▶ the hepatitis virus that damages the hepatocyte, which often increases the liver enzyme ALT).

Autoimmune basics

In autoimmune conditions, there is *no* viral infection. But for some reason (which is still unclear at present), a part of the cell's own protein is placed 'on the stick'. This causes the same chain of events, which leads to the production of a specific antibody. Broadly, for rheumatoid arthritis this Ab is rheumatoid factor (RF); for ankylosing spondylitis (AS) it is HLAb27; for pernicious anaemia it is parietal cell Ab or intrinsic factor Ab; and for systemic lupus erythematosus (SLE) it is anti-nucleic antibody (ANA).

There are hundreds of possible Ab tests, usually undertaken in secondary care by a consultant. If the Ab is present then the patient is called 'seropositive'; if it is not present then the patient is labelled 'seronegative'. For most Ab tests, about 80% of people with the

symptoms are seropositive, leaving 20% seronegative. For these seronegative patients, we could undertake more Ab tests. However, in practice it's more likely that their symptoms will be treated as if they were positive and the effects reviewed. The reason for the existence of symptoms in seronegative patients is unclear.

As discussed in the case study in Chapter 4, autoimmune patients may have blood tests to consider the autoimmune diagnosis (RF), white cell and inflammation response to the condition (WBC, neutrophils, CRP, PV) and any effects of treatments or interventions (RBC, MCV, folate, LFTs and U&Es).

There are also antibodies, called anti-neutrophil cytoplasmic antibodies (ANCA), which 'attack' and then change the role of neutrophils. They are commonly associated with vasculitis, which is the inflammatic destruction of the vasculature; cANCA is associated with granulomatosis with polyangiitis (Wegener's) affecting lungs and kidney; whilst pANCA is associated with glomerular nephritis (◀▶ U&Es). Both types can be associated with ankylosing spondylitis (AS).

Clinical symptoms will drive some of the decision-making in terms of requesting and interpreting autoimmune blood tests. For example, a patient with AS will usually have a different presentation from one with RA. As discussed in Chapter 4, these patients may already have outlying values for WBC, CRP and PV because of their underlying condition. As the condition flares, these changes may be significant but nevertheless clinically expected given the condition. However, we still need to be aware that another new condition (such as a bacterial infection) could also modulate WBC, CRP and PV, so we should question baseline changes and symptoms.

Typical autoimmune profiles

A typical systemic lupus erythematosus (SLE) blood profile could be that about 95% of patients with SLE have a positive ANA. SLE is often seen with thrombocytopenia (low platelets) and high levels of anti-single-stranded DNA Ab (a type of antibody usually found in these conditions, usually requested by a consultant).

A typical RA blood profile could be that about 80% of patients are positive for RF and negative for HLAb27. This often occurs with raised neutrophils and WBC, raised CRP, raised PV and raised globulin.

A typical AS blood profile could be that about 80% of patients are positive for HLAb27 and negative for RF. If a patient is HLAb27 negative then we could consider two new genetic tests: Type 1 tumour necrosis factor receptor shedding aminopeptidase regulator (ARTS1)

and interleukin 23 receptor (IL23R). This often occurs with raised neutrophils, WBC, CRP and erythrocyte sedimentation rate (ESR).

Polymyalgia rheumatica (PMR) means 'pain in many muscles' and is thought to be mainly due to inflammation of the muscle's vascular system. A typical PMR blood profile could be raised ESR and CRP. These patients are usually negative to rheumatoid factor (RF) but positive to HLAb27, and some have elevated platelets and low RBC (although the reason is unclear). If the patient is on certain types of statins, measuring total creatine kinase (CK) may help identify muscle damage. CK is a muscle enzyme and its value in the blood increases in relation to muscle damage. It is thought that some statins can induce muscle damage and cause pain.

Polymyositis is the presentation of muscle antigen as foreign material. A typical blood profile could be positive for ANA and then positive for the Anti Jo subset (a type of antibody usually found in these conditions). These patients could also have raised serum creatinine kinase (CK), and usually raised CRP, PV and ESR.

Reactive arthritis is often linked to an underlying viral or bacterial infection that has induced an immune response and is not subsequently 'switched off'. A typical blood profile could be positive for HLAb27 and negative for RF, with raised CRP and ESR.

Psoriatic arthritis (PA) causes connective and musculo-skeletal damage, which is usually linked to prolonged psoriasis. Psoriasis is the autoimmune destruction of the skin, and a typical PA profile could be negative for RF but positive for HLAb27, with raised CRP and ESR.

Myeloma

A myeloma is a cancer of the B cell lymphocytes. Often, damaged cells aggregate in the bone (◀▶ bone pain, bone profile test and raised calcium (Ca)). The cells often produce a monoclonal globulin protein called a Bence Jones protein. The types of globulins produced by the B cells can be further assessed by means of electrophoresis, which can help differentiate between different types of myeloma and leukaemia. This gives profiles of gamma, IgG, IgM and IgD and so on. The patient will often also have raised WBC (more B cells), raised globulin (from Ab production) and raised PV due to more 'stuff' (globulins and B cells) in the blood and raised ESR.

The correct and normal antibody produced by the B cell has two major components – a heavy chain and a light chain. In some myeloma patients, the faulty B cell produces only the light chain and this is called the Free Light Chain (or FLC or paraprotein or, when measured in the urine, Bence Jones Protein). Take care here, as someone with chronic kidney disease may present with an artificially raised FLC but have no myeloma (their kidney hasn't got rid of the small amount of normal FLCs we make each day). We don't know exactly why this is the case.

Storytelling: Imagine a B cell called Bob. Bob makes a certain type of normal antibody. The clever part is that, once Bob dies, Bob's children will also only ever make the same antibody, and the grand-kids and the great-grandchildren likewise as they are all clones of the original Bob. Imagine we also have B cells called Brenda, Bruno, Bill, Bella, Beatrice, Bellatrix, Bethan and Bomber. Each of these B cells produces a different antibody so we have 'polyclonal' antibody production. All their clones will continue to produce these different antibodies. But if the bone marrow goes wrong and only makes Bobs, then we may switch from a 'polyclonal' profile to a 'monoclonal' profile, as only one type of B cell is being produced.

Fibromyalgia/Chronic fatigue syndrome (CFS)

Fibromyalgia is an example of a condition that significantly affects millions of people and yet we have no single blood test to diagnose fibromyalgic conditions or conditions such as CFS. The current approach is to exclude symptomatic presentation by checking for infection, anaemia, thyroid symptoms, etc., as well as clinical history. The main reason why we have no blood test is due to the likely pathology being the disruption in a neurotransmitter such as substance P, cytokines or chemokines produced by inflammation and the way in which the muscle and nerves talk to the brain. A chemical change is happening but, at present, a simple, cost-effective, reliable test hasn't been identified. Once we have a reliable means of diagnosis, the next challenge will be to find a reliable treatment.

9

Transfusion testing

As blood transfusion involves the transfer of either whole blood or specific blood products between patients, certain screening tests need to be conducted. It may also be appropriate to screen patients' blood to ascertain their blood group or rhesus (Rh) status in case they subsequently require an intervention. Haematology and biochemistry are closely related areas so blood samples for transfusion or blood products might be requested at the same time. Typical tests would include ABO group and Rh type, blood group antibodies, syphilis, HIV and HepB.

The ABO blood group is determined by surface antigens and corresponding antibodies. In A group, the red cell has A antigens and thus anti-B antibodies in the plasma, as the presence of a red blood cell with a B antigen would suggest a foreign or exogenous source. In B group, the red cell has B surface antigens and thus anti-A antibodies in the plasma. For this reason, A blood cannot usually be given to B blood patients. In AB blood group, the red cell has both A and B surface antigens and therefore has *no* antibodies in the blood. Hence, a person with AB can usually receive any blood group. In O blood group, the red cell has *no* surface antigens but usually has both A and B antibodies. Hence, O can usually be given to all groups. As the plasma and red cells contain opposing antigens and antibodies, plasma compatibility is usually the opposite of red cell compatibility.

Rh type denotes the presence or absence of a D antigen, with Rhesus positive (Rh+) having the D antigen present. The Rh type is identified in addition to the antigen status of the red blood cell, for example, O- or AB+. People who are Rhesus negative (Rh-) can usually receive blood from matched ABO Rh- type, whereas Rh+ patients can usually receive either Rh- or Rh+ type.

Whole blood products can also be separated into their component parts: packed red cells from which most of the liquid component (plasma) has been removed; plasma; platelets; and fresh frozen plasma (FFP), plasma that is rapidly frozen which helps to retain key clotting factors.

Hepatitis screening for HepB (HBV) is usually done by checking for the HBsAg or HBcAb, the former being the surface antigen and the latter being the antibody. Some departments may offer the nucleic acid test, which determines the presence of HepB viral RNA. If the patient has an active infection then they are likely to have the antigen and/or the RNA, as well as the antibody (which may take a few weeks to develop). Hepatitis C (HBC) is usually tested by the presence of the HBC antibody.

Storytelling: Thinking about the A blood group in the ABO system, the white cells are your 'protective army' who will destroy foreign invaders. If you clearly mark *your* red blood cells with a *red* flag (A Ag), your army will NOT destroy cells with a red flag. But it WILL destroy cells with a blue flag, using its special anti-blue flag rockets (the anti-B Ab). You can therefore give your blood to other people with red-flagged cells, but not to people with anti-red flag rockets. AB people have red cells with BOTH red and blue flags, so they have NO anti-red or anti-blue rockets and can receive any type of red cell. In O group blood, the red cell has NO flags. O group blood can therefore be given to people in any group, even if they have anti-red or or anti-blue flag rockets. People with O blood can therefore usually only receive type O blood. Hence, type O blood is often referred to as 'universal donor', while type AB blood is a 'universal receiver'.

10

Chronic disease markers: Diabetes

In practice, especially in primary care, patients usually present with one (or both) of the two major types of diabetes: diabetes insipidus (DI) and diabetes mellitus (DM), with resulting polyuria and dehydration.

Diabetes insipidus

Diabetes insipidus is not usually linked to excessive glucose but instead to deficient control of anti-diuretic hormones that control urine output. Investigations to help differentiate DM from DI include: raised glucose and HBa1C, indicating DM; raised Na, suggesting DM and DI; and low vasopressin (antidiuretic hormone (ADH) indicating DI). In both types of diabetes, kidney function may be affected so you should also consider measuring U&Es.

DI has three causes: neurogenic (no vasopressin production in the brain); nephrogenic (kidney does not respond to vasopressin); and dipsogenic (inappropriate thirst mechanism triggered by the hypothalamus, which incorrectly suppresses vasopressin). To differentiate, a desmopressin fluid intake test may be conducted. Alternatively, renal function can be assessed by U&Es (raised Na and urea, and decreased estimated glomerular filtration rate or eGFR), or a thyroid function test can be performed to exclude a tumour (abnormally high or low levels of TRH, TSH and T4). See Chapter 13 on thyroid function tests.

Diabetes mellitus

Diabetes mellitus is a generic name for poor control of glucose. Patients with DM have high levels of glucose (hyperglycaemia) due to a poor insulin response. The main presenting symptom in primary care is polyuria due to osmotic diuresis. This is caused by excessive glucose in the kidney, which cannot be reabsorbed. Osmotic pressure is increased within the tubule, which transiently retains water within the lumen, thus increasing urine output. This subsequently leads to dehydration (polydipsia). There are two main types of DM: Type 1, in which no insulin is produced; and Type 2, in which insulin is not effectively recognised by cell receptors.

Glucose is a type of sugar (a carbohydrate) and provides the primary source of energy in the body, especially in the brain, which is why it needs to be stored effectively. Free glucose, in the blood, lasts a few hours before we become hungry; and stores of glucose last about 16 to 24 hours before hypoglycaemia and confusion occur. Glucose is also highly reactive. It will bind readily to DNA and protein, causing most of the symptoms of diabetes, such as neuropathy, retinopathy and erectile dysfunction. Polyuria is partly caused by destruction of the glomeruli in the kidney and partly because glucose also drives water balance, and thus polydipsia. Hence, there is a vital need to clear excess glucose.

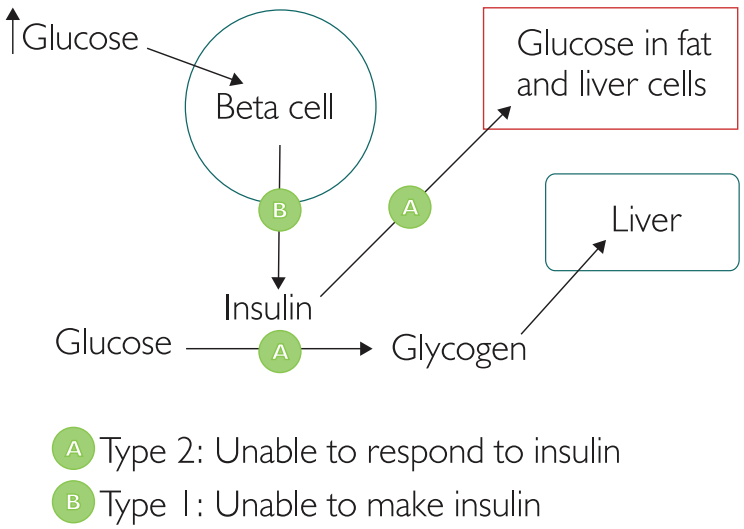


Figure 10.1: Flow diagram of insulin-mediated storage of glucose as glycogen

Glucose causes insulin to be released from beta cells in the pancreas. Insulin converts glucose to glycogen (in a process called glycogenesis), and the glycogen is subsequently stored in the liver. Failure to recognise glucose or make insulin will lead to glucose in excess. Glucose levels can therefore, in a normal state, be affected by stress, pancreatitis and liver damage (◀▶ adrenaline, amylase (raised in pancreatitis) and LFTs, especially following trauma or in patients with poor liver function).

Diabetic complications

Most of the complications in diabetes are caused by excess glucose binding to, and then interfering with the actions of, specialised cells and proteins.

There is raised cardiovascular risk caused by excess glucose binding to the vascular wall. This increases stiffness and oxidises any excess low-density lipoprotein (LDL), which – under the action of macrophages – becomes foam cells and forms atherosclerotic plaques. The glucose also binds directly to the cardiac muscle and nerve endings.

In diabetic neuropathies, the excess glucose binds directly to the nerve cells and suppresses nerve conductivity, inducing vascular and neural retraction from the site. The reduction in vascular flow and integrity leads to more incidents of infection because the white cells are less able to reach the affected site. This is also why antibiotic therapies may take longer to work. This is particularly true in sites with poor circulation such as hands and feet, where neuropathies are most commonly observed in primary care.

In diabetic retinopathy and other vision symptoms, the excess glucose binds directly to the retinal cells and changes their structure. This causes a change in opacity and function, leading to blurred vision and in some cases eventual loss of sight.

In diabetic renal impairment, the excess glucose binds directly to the glomerulus and nephrons, suppressing performance. This often reduces eGFR and increases the already dysfunctional Na levels caused by polyuria and polydipsia.

Diabetic ketoacidosis occurs when fatty acids are utilised for energy, given the poor control of glucose. This produces ketones, which are acidic and affect the blood pH. Investigations should include high blood glucose, presence of ketone bodies, high blood pH, U&E due to renal dysfunction due to continued dehydration, amylase to rule out pancreatitis, and full blood count (FBC) and C-reactive protein (CRP) to rule out infection.

Measuring glucose, and diagnosing and treating diabetes

Glucose levels can be measured in numerous ways, shown A to D in Figure 10.2 (below).

Random blood glucose (A) assesses the patient's ability at a random point to have glucose under control. This is usually used in patients already diagnosed with diabetes, who are managing insulin loads or diet interventions. The random blood glucose can produce false positive values (B), particularly following a meal, especially if the meal is high in sugar. This false positive would return to normal values under the action of insulin over a few hours. Fasted blood glucose (B1) may therefore be requested to remove the chance of a high result being due to a meal.

The oral glucose tolerance test (C) measures the insulin response following a glucose load. The patient attends having fasted and a blood sample is taken (time = 0 min). The patient is then given a glucose load, usually as a drink, which should induce an insulin response. They then wait in a low exercise capacity, for two hours, for the action of biphasic insulin to be

effective. Then a second blood test is taken (time = 120 min). Glucose is measured and compared in both samples; they should be within the reference range in normal patients. In glucose intolerant or diabetic patients, the 120-minute sample will have glucose outside the reference range (C1), highlighting a lack of insulin efficacy.

HbA1c is a type of haemoglobin (HbA) with glucose attached to it (D). Haemoglobin is the oxygen-carrying protein in red blood cells and it becomes irreversibly glycosylated when glucose is in excess. It is therefore a useful long-term marker of diabetes, because the red blood cell has a lifespan of 12 weeks. There are two ways to express HbA1c – either as a percentage or as a concentration. The DCCT percentage values refer to what percentage of total Hb is HbA1c. A value higher than 7% usually indicates glucose intolerance. The IFCC mmol/mol value refers to measuring the amount of HbA1c directly as a concentration. A value higher than 53 mmol/mol usually reflects glucose intolerance.

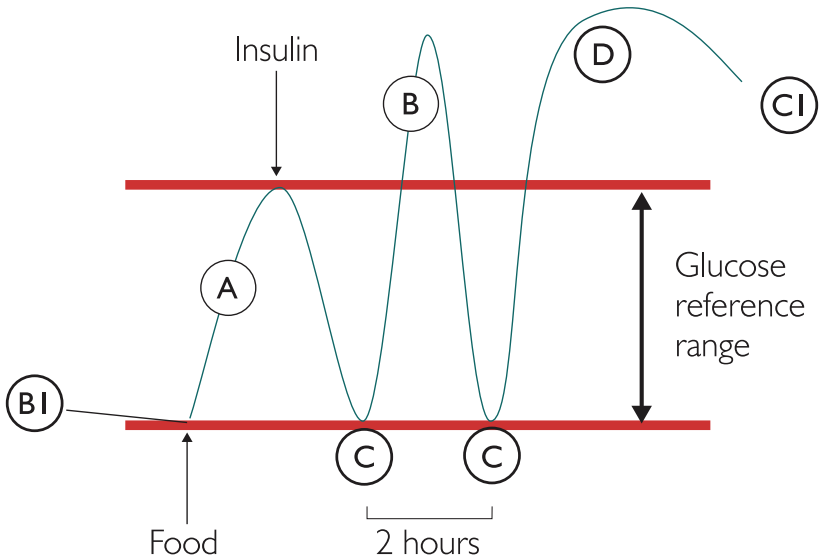


Figure 10.2: Measuring glucose levels

Drugs like metformin (a biguanide class) work by inhibiting hepatic glucose production. This decreases glucose uptake in the gut and increases insulin-sensitive receptors, which increases cellular glucose uptake. Successful treatment with metformin should reduce excess glucose and may also reduce LDL and coronary heart disease risk.

Drugs like exenatide (byetta) work by increasing pancreatic insulin, suppressing glucose transit across the gut and suppressing glucagon (a hormone released to suppress insulin activity via negative feedback).

11

Chronic disease markers: Cholesterol

The risk of developing cardiovascular disease (CVD) can be measured using a variety of tests within the UK NHS Quality and Outcomes Framework (QOF) – for example, the percentage of patients with coronary heart disease (CHD) whose last measured total cholesterol (measured in the previous 15 months) is 5mmol/l or less.

Wider lifestyle choices could also be addressed with full blood count (FBC), liver function tests (LFTs) and cholesterol measures. Red blood cell count (RBC) may be increased in response to tissue hypoxia caused by smoking (polycythaemia), which may elevate plasma viscosity (PV) and increase clot risk. The presence of plasma cotinine can be used as a marker of nicotine exposure in an active smoker. Homocysteine is an amino acid that causes arterial stiffness, and is present in folate deficiency (often caused by the poor diet seen in smokers and alcoholics). Other tests would include an increased gamma-glutamyl transferase (GGT), which would also suggest alcohol intake (see LFTs, Chapter 16). Total cholesterol should be checked as a CHD marker, using HbA1c or fasting glucose to address any underlying diabetes. Finally, there may be a link to hypothyroidism, which should be investigated with a thyroid function test. A genetic condition called familial hypercholesterolaemia should also be checked as appropriate.

'Good' and 'bad' cholesterol

Cholesterol has several functions: development of cell membranes; production of bile (which makes fats soluble and absorbs vitamins); and production of vitamin D and steroids (cortisol and aldosterone) and sex hormones (progesterone, oestrogen and testosterone). Cortisol releases glucose in response to stress, and aldosterone increases blood pressure. These are evolutionary fight-or-flight mechanisms, *but* high stress, glucose and high blood pressure are all lifestyle risks in CVD.

As cholesterol is fat-soluble and is needed by various cells, it is transported in the water-based blood by chylomicrons. 'Bad' low-density lipoprotein (LDL) and 'good' high-density lipoprotein (HDL) are types of chylomicrons. **Remember that LDL could be known as Lousy and HDL as Happy.**

Storytelling: In LDL, the 'L' can be called 'lousy' as it's bad, and the 'H' in HDL can be called 'happy' as it's good. Total cholesterol is like 'all the people invited to a party'. It's all the cholesterol in the blood – good, bad and ugly. A helpful marker is the cholesterol ratio, which the UK Q-Risk 3 system sets at 4 and below. If you are planning a party, it's just as important to think about WHO to invite as HOW MANY to invite. According to the ratio, every 1 happy person (HDL) will counter the effect of inviting up to 4 grumpy or lousy people (LDL).

Cardiovascular risk

Total cholesterol is usually measured with the patient having fasted. It is a complex equation, which takes into account LDL cholesterol, HDL cholesterol and triglycerides. The National Institute for Health and Clinical Excellence (NICE) and Department of Health cholesterol guidelines specify a normal/healthy threshold of total cholesterol less than 5.0mmol/l and LDL cholesterol less than 3.0mmol/l.

HDL transports cholesterol from the arterial wall and blood to the liver (to be excreted) and adrenal glands (to be used to make cortisol). In this way, HDL helps to reduce cholesterol levels and CHD risk and may also modulate CVD risk by directly inhibiting the LDL-induced inflammation and platelet aggregation in the vasculature.

Triglycerides

Triglycerides (TGs) are the fat equivalent of glycogen. They are stored fat molecules. Some TGs are transported in the blood between adipose tissue and muscle and therefore reflect a high-fat diet. There is a relationship between high TGs and low HDL, although the reasons why a high TG level is a risk factor for CVD are not fully understood. One hypothesis is that the 'type of fat' (such as omega 3) being incorporated into the vascular wall may be more (or less) susceptible to being oxidised. This may lead to the vascular wall becoming more (or less) structurally 'stiff'. If the vascular wall becomes less able to flex, this increases CHD risk.

Cholesterol treatments

Statins reduce cholesterol/LDL production in the liver by partly suppressing the enzymes HMG-CoA reductase and Acetyl-CoA. **Storytelling: These enzymes help to load the logs onto the ferry in the liver in order to deliver the cholesterol to the vascular walls.** By suppressing this activity, the cholesterol delivery (total cholesterol in the blood)

is reduced. A side effect in some patients is the separate induction of muscle destruction, leading to muscle pain. Some practices may simply choose to change the statins, but in some cases it may be appropriate to measure muscle damage. A helpful test for this is total creatine kinase (CK). Muscle pain with creatine kinase (CK) levels more than ten times the upper limit of normal (ULN) may indicate clinically important myositis and rhabdomyolysis. If pain continues following statin withdrawal, an additional pathology may be present.

Cholesterol is usually used as a predictor of CHD. Another predictive cardiac marker is b-type natriuretic peptide (BNP). An increase in BNP correlates with increased cardiac wall load and dysfunction.

Following a cardiac event, endogenous cardiac muscle proteins or enzymes (due to the muscle damage) are present in the blood. CKmb is a type of creatine kinase found in heart tissue, which is released into the blood following cardiac damage, making it a useful indicator of a cardiac event. CK muscle brain (CKmb) rises after three hours, and myoglobin rises after seven hours. Aspartate aminotransferase (AST) may rise after two days. (AST is a liver enzyme, but it is also found in the heart – ◀▶ normal other LFT values.)

Troponins peak after seven hours and remain elevated for up to seven days. Troponins are muscle structure (contraction) proteins, which are released into the blood after muscle damage. Troponin I and C are markers of cardiac muscle damage, which is commonly used to differentiate between an angina and a myocardial infarction (MI). MI often results in significant troponin I release compared to angina, although care is needed when making a final interpretation, as some non-myocardial events (like tachycardia) can raise troponins. Patients with chronic obstructive pulmonary disease (COPD) may also have increased levels of troponins, given the ischaemic and hypoxic nature of the disorder.

12

Chronic disease markers:
Chronic obstructive pulmonary disease and acid base

Chronic obstructive pulmonary disorder (COPD) is a narrowing of the airways, leading to poor lung function and severe airflow obstruction (low FEV_1). The cause of COPD is not clear, although there are several contributing factors such as smoking, exposure to workplace dust and particulates, air pollution and genetic factors (in about 2% of cases). It can also partly result from an autoimmune condition following prolonged inflammation, as seen in other autoimmune conditions.

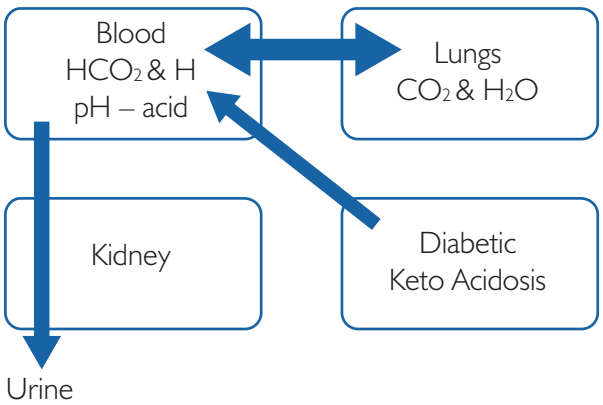


Figure 12.1: COPD Interactions

Patients with COPD may be more susceptible to upper and lower pulmonary infections – hence the importance of vaccinations. White blood cell count (WBC) may be a helpful marker for infection, as would the presence of a functional antibody (Ab) following vaccination.

Given the restriction in oxygen caused by COPD, the patient may develop polycythaemia with a raised red blood cell count (RBC), especially if they also smoke. However, in practice, in an elderly patient, the hypoxia-induced production of erythropoietin (EPO) may be mitigated by poor renal function (with an estimated glomerular filtration rate <60) and raised RBC may not occur, as EPO is made by the kidney.

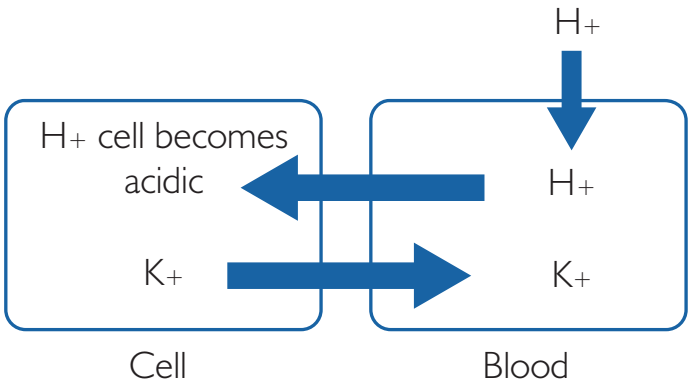


Figure 12.2: Consequences of Acidosis

In addition, tuberculosis may need to be excluded in COPD patients. It may also be appropriate to screen for alpha-1-antitrypsin, which provides important protection from attack by white cells. White cells, particularly neutrophils, use potent enzymes to destroy bacteria. To protect our own cells from attack, we make alpha-1-antitrypsin, which can block or inhibit these enzymes. In some autoimmune conditions and COPD, the patient may have a deficiency or lack of alpha-1-antitrypsin and the raised neutrophils can therefore cause more damage.

Acid base measures

Acid base measures are helpful in COPD patients, as well as in those with diabetes or drug overdose and those taking proton pump inhibitors. Acid and alkaline conditions are determined by the concentration of H ions (protons). A high H^+ concentration is a low acid pH, and a low H^+ concentration is a high alkaline pH. Most enzymes work within a narrow range of pH (around 7.4) to control these two systems, metabolic and respiratory, using the kidney and lungs respectively to ensure that this pH is maintained. Changes in H^+ concentrations are controlled by buffers.

The equation

$$H^+ + HCO_3^- \text{ (metabolic)} \leftrightarrows CO_2 + H_2O \text{ (respiratory)}$$

is used to describe the bicarbonate (HCO_3) buffer system. Thus, if the pH rises (more H^+), this moves the equation to the right, producing more CO_2, which is cleared by a subsequent increase in ventilation. Equally, a poor ventilation rate will increase CO_2 and move the equation to the left and increase pH. This is why COPD patients often have respiratory acidosis.

These compensatory mechanisms also function within a particular timespan. Physiological buffers, such as the bicarbonate–carbonic acid buffering system, work immediately. Pulmonary compensation occurs next, within a few minutes. Finally, about six hours after sustained acidosis or alkalosis, renal compensation occurs. In certain patients, bone can also be used as a last resort, as it contains high levels of bicarbonates, although this disrupts bone density. Therefore patients with COPD, renal dysfunction (chronic kidney disease) and bone dysfunction (osteoporosis) may have a very limited buffering capacity and will need to be carefully assessed.

Acid base derangements

The most common acid base derangements are metabolic acidosis, metabolic alkalosis, respiratory acidosis and respiratory alkalosis.

In metabolic acidosis, the pH is low, meaning high H^+ load. Causes of metabolic acidosis include lactic acidosis, diabetic ketoacidosis (ketone bodies are produced from fat, which are acidic), and loss of bicarbonate through severe diarrhoea or bicarbonate wasting through the kidneys or gastrointestinal (GI) tract. This is metabolic, partly because cells are producing excess acid and partly because the kidney attempts to clear H^+. H^+ in excess can move into the cells and cause K^+ to be shunted out, leading to hyperkalaemia and a possible coronary heart disease event.

As HCO_3^- increases (usually as the result of excessive loss of metabolic acids), metabolic alkalosis occurs. Causes of metabolic alkalosis include Cushing's syndrome, some diuretics, secretory adenoma of the colon, and exogenous steroids.

Respiratory acidosis (pH <7.35, $PaCO_2$ >45 mm Hg) reflects alveolar hypoventilation. Given that the renal control of HCO_3^- is tightly controlled, large and prolonged changes of $PaCO_2$ are required to increase pH. As discussed, this is usually seen in primary care as COPD, and in acute care as brainstem injury from acute ingestion of opioids. Supportive O_2 treatments may be used, or in acute cases of overdose corrective therapies like intravenous naloxone may be considered.

Respiratory alkalosis usually occurs as a result of hyperventilation, often caused by mechanical over-ventilation, hepatic disease, 'panic attacks', pregnancy and septicaemia. Treatments are usually corrective, such as controlling breathing. However, the patient should be monitored to prevent subsequent metabolic acidosis.

Table 12.1: Sample acid base results

pH	pCO_2	HCO_3^-	Interpretation	Example
HIGH	LOW	NORMAL	Respiratory alkalosis	Hyperventilation
LOW	HIGH	NORMAL	Respiratory acidosis	COPD
HIGH	NORMAL	HIGH	Metabolic alkalosis (MAL)	Cushing's, Diuretics
LOW	NORMAL	LOW	Metabolic acidosis (MAC)	Diabetic ketoacidosis

It is also possible to have a mixed acid base. For example, a patient who has lactic acidosis (metabolic) and COPD (respiratory) might have the following results: pH 7.12, CO_2 55, HCO_3^- 14, and so on.

13

Thyroid function

The thyroid is a gland in the neck, which produces thyroxine, which ultimately modulates cellular energy expenditure. A thyroid function test (TFT) can be complex to interpret but most hospital departments will be able to provide detailed interpretations. A TFT has three main purposes: to diagnose problems with the thyroid (and pituitary/hypothalamus axis); to differentiate from another condition (such as pernicious anaemia, which also raises anti-thyroid antibodies); and to monitor the titration of thyroxine (T4) treatment.

In simple terms, environmental factors such as light and heat, signals from dietary status, and stress and depression send information to the hypothalamus, which produces thyrotropin-releasing hormone (TRH). TRH acts on the pituitary gland to produce thyroid-stimulating hormone or thyrotropin (TSH), which elicits thyroxine (T4) from the thyroid gland. Elevated levels of T4 can act as a negative feedback to switch off TRH production. In clinical practice, T4 will be measured if TSH is out of range.

T3

T3 is produced from T4 and is a more biologically active thyroid hormone. In practice, since T3 is derived from T4, T4 is usually used to monitor thyroid function. However T3 can be requested if the TFT values are deranged, and further investigation is needed. T3 is helpful in the early diagnosis of hyperthyroidism (see below), as T3 rises before T4. The main uses of T3 are to monitor T3 therapy and to address hyperthyroidism in a patient with a low TSH. Remember, hyperthyroidism should usually have a high TSH. A low TSH could therefore indicate a non-thyroidal illness (NTI). NTI is usually seen in acute conditions, such as diabetic keto-acidosis, after an MI, in severe starvation and in critical patients in the intensive therapy unit (ITU). It is thought to be due to a dysfunctional thyroid pituitary hypothalamus feedback loop (see Figure 13.1 below).

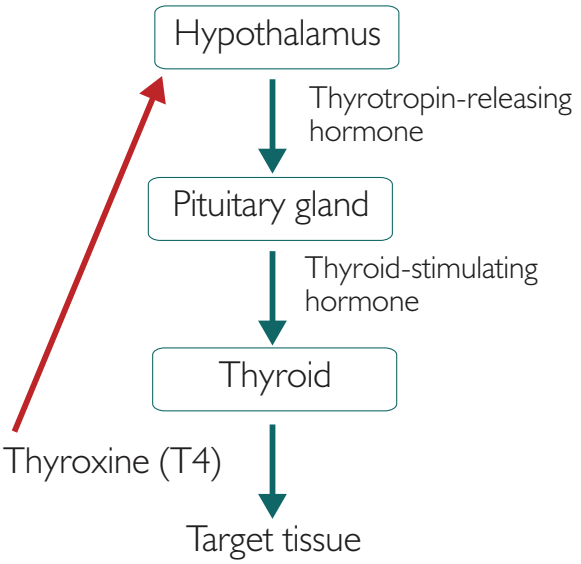


Figure 13.1: Thyroid hormones

Hyperthyroidism

Hyperthyroidism is raised T4, leading to increased metabolic rate (often with a goitre), agitation and weight loss. Most cases are primary Grave's autoimmune, although a rare secondary form caused by a tumour of the pituitary overproducing TSH also exists. It is often treated with anti-thyroid drugs (carbimazole), iodine and beta blockers. A primary condition is when the thyroid alone is affected. In a secondary condition, the pituitary is affected and over- or under-produces TSH, which in turn affects the thyroid.

Hypothyroidism

Hypothyroidism is low thyroid production, often with a reduced basal metabolic rate and weight gain. Most cases are primary Hashimoto's autoimmune, which destroys the thyroid tissue. As discussed earlier, some other autoimmune conditions, such as pernicious anaemia, raise auto-antibodies against the thyroid. Physical and chemical damage from trauma and radiation, as well as inappropriate sex hormone signals (sometimes seen in endometriosis) can also reduce T4 production.

Table 13.1: Sample thyroid function test results

TRH Hypothalamus	TSH Pituitary	T4 Thyroid	Observation
Normal	Normal	Low	Primary
Normal	Low	Low	Secondary
Low	Low	Low	Tertiary

Thyroid treatments

T4 medication can take between four and six weeks to affect and stabilise TSH, and this would be the optimum time after which to repeat the TFT. If too much T4 is given, this will suppress the hypothalamus, and free T4 after six weeks will be very low. The usual range for T4 is 12.0–23.0pmol/L, and for TSH it is 0.4–5.0mU/L. Therefore a TSH of under 0.01mU/L usually indicates over-replacement; and 0.01–0.4 mU/L may mean some over-replacement, especially if free T4 is >30mU/L. A TSH of 0.4–5.0mU/L usually means that the T4 replacement is sufficient, whilst a TSH of >5.0mU/L indicates probable under-replacement or patient non-compliance. This is due to the pituitary still producing high amounts of TSH to encourage the thyroid to make T4 (which it won't).

Since T4 can take up to six weeks to stabilise an adequate TSH response, patients newly commenced on thyroxine could have a repeat TFT around six to eight weeks post-intervention. Once the dose is stable, following a round of six-monthly TFT, you can then consider (if clinically appropriate) moving to an annual TFT.

Drugs such as carbimazole, normally prescribed to suppress T4 production, can be used to 'block' free T4, or they can be used as a therapy to 'block' before medicating with thyroxine to 'replace'. As with T4 supplementation for hypo-conditions, TSH levels in patients with hyper-conditions may take up to 12 weeks to respond to carbimazole. In practice, you should consider carrying out a TFT every four to six weeks, then move to every three months once stable. For other treatments, such as radioiodine, consider an interval of four to eight weeks after dose, but take clinical advice in your own healthcare setting.

14

Bone profile

The bone provides skeletal structure, bone marrow production of cells, and over 99% of stored calcium (Ca). Bone is made of two structural components: the cortical (compact) and cancellous (trabecular). The cortical is the hard outer layer, making up about 80% of the bone mass. The remaining mass is mainly the trabecular, which contains blood vessels, cells and bone marrow (which makes red cells, white cells and platelets). In addition to the blood cells produced by bone marrow, bone also has two major types of cells – osteoblasts and osteoclasts. Osteoblasts mineralise bone (adding to it), whereas osteoclasts break down bone. There is another cell called an osteocyte, which is thought to modulate calcium concentration and respond to load bearing, and possibly exhibit phagocytosis.

A typical bone profile will measure calcium, phosphate, albumin, alkaline phosphatase and in some cases vitamin D and parathyroid hormone (PTH).

Bone turnover and osteoporosis

In normal bone turnover (see Figure 14.1), the osteoclast (A) liberates Ca from the bone. This Ca is then used for metabolic processes, and over a few hours Ca levels fall. In response to this, the parathyroid gland (PTG) produces PTH and the kidney releases calcitriol (Vit D_3), originally derived from vitamin D in diet and skin via exposure to ultraviolet (UV) rays in the sunlight. Given the key role of the kidney-derived vitamin D_3 in this process (◀▶ urea and electrolytes or U&Es), a patient with dysfunctional renal output (as shown by a reduced estimated glomerular filtration rate (eGFR) of less than 60) may be less able to resolve a fracture and may be more predisposed to bone conditions. The PTH/VitD_3 complex induces increased osteoclast activity in the bone, enhances Ca uptake in the gut and increases reabsorption of Ca in the kidney. The increased calcium (as a result of the PTH/VitD_3 complex) is reabsorbed into the bone by the osteoblasts (B). The speed and frequency of this process is partly suppressed by the action of the hormone oestrogen (green circle).

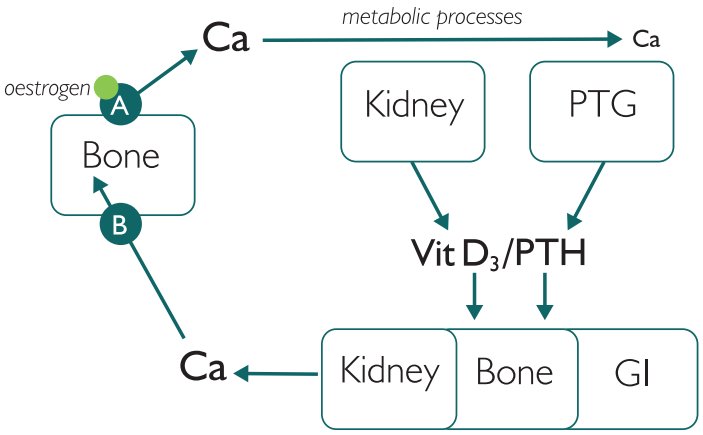


Figure 14.1: Normal bone turnover

In post-menopausal patients or those receiving long-term anti-oestrogen/progesterone treatments, this process is less regulated and more likely to result in frequent fractures, as the bone remodelling increases. This condition is called osteoporosis, and initial pre-menopausal bone mineral density (which is in turn controlled by diet, lifestyle and genetics) is an important factor in disease risk and progression. As the process is speeded up, variations in the blood components are difficult to observe. Thus, bone profile is not usually used in practice to diagnose or monitor osteoporosis. In elderly patients with chronic kidney disease, osteoporosis can be exacerbated. In addition to not being able to sustain $VitD_3$ production, these individuals have compromised reabsorption of Ca, which increases their risk of fracture and the time needed for any fractures to heal.

Paget's disease

In Paget's disease there is a dysfunctional regulation of the osteoclast and osteoblast pathway. This leads to imperfect removal and reabsorption of Ca.

Storytelling: Imagine repairing a road. Team A (osteoclasts) remove two buckets of rubble (Ca) from the road (bone) and dispose of the Ca by using it to make muscle, nerves and teeth. But they tell Team B that they have taken out 20 buckets of rubble. The foreman (PTH) therefore insists that Team B (osteoblasts) use 20 buckets to refill the hole (even though there is only room for two buckets of rubble). Thus, the road will not return to its original flat surface. Instead, a bump will be left.

In practice, this process leads to bone protrusions and pain. A raised Ca level is sometimes seen in patients with immobility due to joint degradation, as the sockets and joints are ill formed.

Drugs like bisphosphonates work by binding to calcium in the bone, which then suppresses osteoclast activity, thus improving and maintaining bone mineral density. Bisphosphonates are therefore often used to treat osteoporosis and Paget's.

Osteomalacia

Patients with a vitamin D deficiency can be predisposed to osteomalacia, known as rickets in children. Vitamin D is obtained from the diet and stored in the skin. There, under the action of UV light, it is converted into vitamin D_2 (calcidiol), which is stored in the liver. Vitamin D_2 is then transported to the kidney, where it is converted, by a parathyroid hormone (PTH) induced enzyme, into vitamin D_3 (calcitriol 1,25 $(OH)_2D$). Inadequate production of vitamin D_3 often results in hypocalcaemia, leading to dysfunctional bone remodelling and eventually weak bones that 'bend' under the weight of the trunk. Causes of vitamin D_3 deficiency may be a lack of UV light, too little vitamin D in the diet, or the presence of chemicals in the diet that can block vitamin D_3 absorption in the gut (seek advice from dietetics), or renal impairment as in chronic kidney disease. Blood test results in osteomalacia are often low Ca, high PTH, as the parathyroid attempts to correct the low Ca, and low phosphate, as PTH increases excretion in the urine (◀▶ eGFR for renal status).

Magnesium

Other tests that may be returned are magnesium (Mg), alkaline phosphatase and phosphate. Magnesium is required to produce PTH so a magnesium deficiency will restrict PTH and thus the availability of free Ca. Hypomagnesaemia is common in alcoholic patients, due to malnutrition, diarrhoea and increased excretion; the latter is also seen in diabetic keto-acidosis. Some thiazide and loop diuretics, antibiotics, proton pump inhibitors, pancreatitis and gastrointestinal (GI) disorders (such as Crohn's) may also lead to low Mg levels.

Alkaline phosphatase

Alkaline phosphatase (Alk Phos) is an enzyme found within the bone, liver, kidney and placenta and is a helpful differential. For example, if a patient has a raised Alk Phos under the LFT, but all other LFT values are normal (as are the U&Es), this could suggest that the cause of the raised Alk Phos could be bone – especially with a raised Ca.

Phosphate

Phosphate forms a complex with Ca to provide the structure of bone. Phosphate levels are mainly controlled by renal output and impairment. Low levels can be a red flag, as this can lead to respiratory failure – given the control of muscle by Ca and Phosphate.

Calcium

Whilst 99% of calcium is stored in the bone, the remaining 1% is bound to albumin as a transport protein and free, biologically active Ca. Therefore, a low albumin level, as in malnutrition or liver damage (◀▶ LFT), may cause an artificially low Ca. Hence, 'corrected' calcium is used, which corrects the Ca for the levels of albumin. Generally, corrected Ca is used in clinical practice. Ca levels are controlled by osteoclasts, osteoblasts, parathyroid hormone, vitamin D (from UV light and the kidney) and the negative feedback hormone calcitonin. Bone profiles are helpful in order to assess some dysfunctional bone conditions such as osteomalacia; as a differential of bone pain as seen in myeloma; and to assist in determining a secondary bone metastasis (bony mets), which would be an unregulated increase in Ca.

Raised calcium (hypercalcaemia) may be caused by a primary hyperparathyroidism, a tumour secreting a PTH-like compound (for all PTH investigations, these are diurnal, so check the timing of the test), over-supplementation of calcium and vitamin D, renal failure, sarcoidosis, myeloma, bone fractures and bone cancer (primary and secondary). It may also be due to inappropriate blood collection.

Low calcium (hypocalcaemia) can therefore be due to low albumin, vitamin D deficiency, renal failure, or damage to the parathyroid gland as is sometimes seen after thyroid surgery.

15

Renal function: Urea and electrolytes

The kidney has three key functions: regulating blood volume, urine and electrolyte concentration (via reabsorption and excretion); the endocrine production of erythropoietin and vitamin D_3; and the excretion of metabolic breakdown products such as urea and creatinine.

The abbreviation U&Es stands for urea and electrolytes. A typical U&E panel may include sodium (Na), potassium (K), urea, creatinine, estimated glomerular filtration rate (eGFR) and uric acid (urate). The U&E blood test is often used to determine renal function. In this chapter, patient case studies are used to explore and interpret some common results.

Sodium

Sodium is an electrolyte that is generally found outside cells in the blood. It controls water balance. **Storytelling: Like putting salt on a slug, sodium draws water out by means of osmosis.** Sodium is linked to the renin–angiotensin system in the following equation:

Na concentration = mass/volume

Mass consists of Na *in* (diet), minus sodium *out* (sweat, excretion). Volume is water *in* (liquid and food), minus water *out* (urine).

In this section, sample patients will be used to describe electrolyte imbalances in various conditions – some common and some very rare.

Hypernatraemia (raised Na)

Patient 1 is hypovolemic, with low water intake, increased sweating, polyuria or diarrhoea.

Patient 2 is euvolemic, with increased water excretion, as seen in diabetes.

Patient 3 has hypervolemia, due to having ingested large amounts of salt, either through diet or accidentally from drowning in seawater (although you are unlikely to see the latter in primary care!).

Patient 4 has Conn's syndrome or Cushing's disease and is being treated with mineralocorticoids (hormones that control water and sodium), with poor patient compliance or incorrect dosages.

In elderly patients, Na intake from the diet may be high, water intake may be low and they may also have chronic kidney disease, so hypernatraemia may be more likely, especially if GFR is <60ml/min and urea levels alone are high. As discussed earlier, the values for eGFR, Na and urea may be abnormal but you may decide (in primary care) to view these results as not clinically relevant in elderly, dehydrated patients. Clearly, any prolonged dehydration can be critical and a red flag.

Hyponatraemia (Na <120mmol/L)

Patient 1 has hyponatraemia and oedema that could be due to congestive heart failure, liver failure (changes in blood flow or volume due to cirrhosis, which leads to inappropriate antidiuretic hormone (ADH) secretion) or nephrotic syndrome ('leaky kidneys' that cannot control excretion products). It may also be linked to chronic obstructive pulmonary disease (COPD) – see Patients 3 and 4.

Patient 2 has hyponatraemia and dehydration due to diarrhoea or Addison's disease. Addison's disease is a condition that affects the adrenal gland and thus blood pressure and glucocorticoid levels. These symptoms may also be seen in renal salt wasting. Renal salt wasting is often due to a trauma, tumour or haematoma in the brain. It may present with similar symptoms to the syndrome of inappropriate anti-diuretic hormone (SIADH). However, Na levels are often greater in SIADH than in cerebral salt wasting syndrome (CSWS), especially when fluid is restricted.

Patient 3 has inappropriate (vasopressin) ADH secretion, which could be due to drug treatments, central nervous system disease or a chest disease such as COPD.

Patient 4 has COPD and hyponatraemia. Usually, the more severe the COPD, the more severe the hyponatraemia, which is then a risk factor for pneumonia. The link, as in Patient 3, is usually due to hypercapnia (increased CO_2), which affects the renin, angiotensin, aldosterone, atrial natriuretic peptide and vasopressin (ADH) pathway, reducing renal blood flow. (See Chapter 12 on acid base, which discusses the link between the pulmonary and renal systems.) This results in water retention, oedema and hyponatraemia. However, in some very rare cases, this may be masking a pituitary adenoma or similar tumour, especially if the hyponatraemia persists as the COPD improves. Consulting the patient history, intervention with medication then retesting, MRI scans and hormone screening (for thyroid-stimulating hormone or thyrotropin and others) may be helpful in such cases.

Patient 5 has hyponatraemia and normal volume, which could be from excess water intake following diarrhoea and vomiting. Some diuretic therapies and the use of ACE inhibitors can cause hyponatraemia.

Potassium

Potassium (K) is an intracellular electrolyte that is mainly found inside the cells. It controls calcium channels by affecting the electric potential difference across the cell membrane. This process is critical for muscle and nerve integrity. A significantly elevated concentration of potassium in the blood is a potential red flag, as it can interact with the cardiac muscle, leading to a cardiac event.

Potassium controls cellular production of hormones such as insulin. It also partly regulates H^+ ions by maintaining a positive charge within the cell, keeping H^+ ions out and thus controlling pH. (◀▶ potassium to acid base.) Potassium levels are tightly controlled by the kidneys.

Hypokalaemia (K <3.5mmol/L)

Patient 1 has been affected by inappropriate use or dosage of diuretics, although most are now K sparing.

Patient 2 has severe, acute diarrhoea and vomiting.

Patient 3 is an elderly patient, living alone, with a very poor diet, who also enjoys liquorice. Some types of liquorice contain mineralocorticoid-type compounds that act on the renal tubules and retain sodium, and thus water. This can lead to hypertension and oedema, and can also increase excretion of potassium.

Patient 4 has had an ileostomy.

Patient 5 abuses laxatives due to bulimia, and also induces vomiting.

Hyperkalaemia (K >5.5mmol/L)

Patient 1 had blood taken using an over-tight and prolonged tourniquet. The blood was then syringed into a vacutainer, shaken and placed in a fridge. The raised potassium is probably a result of this poor phlebotomy technique, and represents the muscular damage at the draw site and the subsequent red cell lysis in the tube (haemolysis).

Patient 2 has had a muscular crush injury and the rise in potassium is due to cellular destruction. The patient is also likely to have raised CKmm (muscular skeletal) and may need to be monitored for a subsequent cardiac event.

Patient 3 has an acidosis, such as diabetic ketoacidosis. This leads to an increase in H^+ ions, which enter the cell and displace K^+ into the blood.

Patient 4 is taking potassium-sparing diuretics or KCl supplements.

Patient 5 has renal dysfunction and thus cannot excrete potassium effectively.

Patient 6 is taking ACE inhibitors, ibuprofen and an antibiotic. All of these could interfere with the urinary excretion of potassium.

Patient 7 has a mineralocorticoid deficiency, such as Addison's disease.

For all of the above patients, additional tests such as HbA1c (diabetes), pH, bicarbonate (acid base), hormone screening (adrenal, pituitary), Na, Urea (renal), LFTs (liver), full blood count (sickle cell and other blood disorders), as well as MRI scans, ECGs and patient histories, can help differentiate.

One of the treatments for acute hyperkalaemia involves shunting the potassium back into the cells. This can be done by using a dose of insulin, bicarbonate or a β2-selective catecholamine (such as salbutamol), as clinically appropriate.

Whilst Na and K are helpful in determining electrolyte and water balance, urea and creatinine are useful in order to specify a pre-, true and post-renal location, and to assess acute versus chronic conditions.

Urea and creatinine

The kidney is very sensitive to urea and actively excretes it into the urine. We have a lot of urea so when the kidney is dysfunctional, urea tends to rise rapidly in the blood (see p. 22). Given that we have much more urea than creatinine in our blood, we wouldn't expect creatinine levels to increase rapidly in a short period of say five days. If it does, this could be serious and the creatinine level is measured by the Acute Kidney Injury (AKI) blood report.

Creatinine is a product of muscle turnover and is a marker of chronic renal failure, demonstrating prolonged damage to the nephrons. A renal stone or a prolonged urinary tract infection (UTI) may slightly increase creatinine over time.

Urea is produced from the breakdown of protein, and is cleared via the urea cycle, which controls nitrogen stores in the body. In practice, it is commonly used as a marker of acute renal dysfunction. As discussed earlier, urea can rise sharply in acute dehydration, and also as an artefact following the intake of a high protein meal. Urea levels rise in acute renal dysfunction due to renal stones, viral infection and prostate cancer.

Patient 1 is a 70-year-old man with a history of lower back pain and infrequent urine production. He is almost anuriac. His urea, Na, and Alk Phos were all significantly raised. This could have been due to a renal stone, or UTI. Dehydration is unlikely, given the increased Alk Phos. However, a significantly increased prostate specific antigen (PSA) revealed

an underlying prostatic tumour, with raised Ca and Alk Phos results from a bone profile, suggesting bone metastasis. On initial presentation to primary care, given that the patient had had a prostatectomy, prostate cancer was not thought to be likely.

Patient 2 is a 35-year-old woman with lower back, pelvic and abdominal pain. Her U&Es were normal, so CA-125 was performed. CA-125 is highly correlated with ovarian cancer, with about 80% accuracy. It is also linked to very severe endometriosis. Following a laparoscopy, this was indeed diagnosed.

The urea:creatinine ratio can commonly be used to determine the site of renal failure and for suspected gastric bleeds. To work this out, you should first take the median (middle) value of the reference range for both urea and creatinine. In this example, the units have been standardised to μmol/L, the range of urea is 3000–8300 and creatinine is 40–130. The medians are therefore 5650 for urea and 85 for creatinine, which is a baseline value of 66:1. To help remember the distinction between creatine and creatinine, think about the test for kidney function, U&E or Ewes and Knees; this has an 'n' sound, as does creat*in*ine (found in the kidney).

In pre-renal dysfunction, such as arterial stenosis, congestive heart failure or dehydration, urea reabsorption is increased and the U:C ratio will favour urea $<$100:1. A U:C of 40–100:1 (normal) is usually seen in post-renal obstructions. In true renal damage, urea reabsorption is compromised and the U:C ratio will be 1–40:1.

Patient 3 is a child with an upper gastrointestinal (GI) bleed and an expected U:C may be 30–40:1.

The overall renal function can be monitored by testing estimated glomerular filtration rate (eGFR). The rate is 'estimated' because, unlike an actual GFR (which involves urine collection), eGFR is measured using only blood. The glomerulus is a structure within the kidney, which connects the vasculature and renal architecture and provides an initial filter for large proteins and cells. **Storytelling: The glomerulus is a bit like a sieve filled with cotton wool, under a running tap.**

A normal eGFR is around 100ml/min. But age, even without specific disease pathology, affects the glomerulus. From the age of about 35, the eGFR value falls by about 10% per decade. It may therefore be 'normal' for an 80-year-old patient to have an 'abnormal' eGFR of 60. However, an eGFR of 60 in a 19-year-old would be more worrying. Since eGFR is based on creatinine clearance (and thus muscle turnover), patients with an African or Caribbean heritage, or with a large muscle mass, should consider having their eGFR adjusted by multiplying by 1.2.

Glomerular filtration rate

The eGFR forms the basis for chronic kidney disease (CKD) staging and sets the scene for a differential or adjunct diagnosis where renal function can affect the pathology, such as COPD, acid base, anaemia, vitamin D deficiency, diabetes and so on. In clinical practice, an eGFR >60ml/min is usually adequate. Some laboratories will therefore not return a numerical value if greater than this and simply report eGFR >60. Some may give numerical values, and more commonly a narrative about CKD status. However, there is some debate about the clinical relevance of some mild staging, particularly in the elderly, for the reasons discussed above.

Table 15.1: CKD stages and eGFR values

Stage	GFR
Stage 1 with normal or high GFR	>90ml/min
Stage 2 Mild CKD	60–89ml/min
Stage 3A Moderate CKD	45–59ml/min
Stage 3B Moderate CKD	30–44ml/min
Stage 4 Severe CKD	15–29ml/min
Stage 5 End Stage CKD	<15ml/min

Urate and gout

White blood cells contain uric acid. When a white blood cell is broken down at the end of its lifespan, uric acid is released into the blood. The uric acid is then removed by the kidneys. If uric acid levels in the blood get too high (either due to excessive input or insufficient removal), the uric acid may form a crystalline structure in the joint and the patient may present with gout. Interestingly, if the patient has gout, a blood test for uric acid may produce a normal result. This is because the uric acid is not in the blood any more – it's now in the joint, in the form of gout. We should therefore measure uric acid levels 4–6 weeks after the patient develops gout. Urate or uric acid is often raised in patients with gout.

Urate is a breakdown product of cellular metabolism, and – more specifically – DNA breakdown. Urate is held in the blood as soluble crystal. However, if the levels of urate rise or a renal impairment affects blood volume and flow (or both), it will quickly precipitate out of solution and form a solid crystal, usually at the interphalangeal joints of the toes.

Storytelling: Imagine floating sticks down a shallow river that is flowing over rocks. The more sticks (urate) you throw in, the more likely they are to get stuck. Also, if the level of the river (blood volume or flow) falls, the sticks will get stuck amongst the rocks.

Urate levels can be raised through poor diet, chemicals called purines and eating food high in DNA such as liver and pâté. High urate may also be seen in leukaemia patients due to the high DNA turnover in the breakdown of large numbers of white blood cells. (◀▶ gout to FBC: WBC.) However, in view of the inflammation, a rise in white blood cell count (WBC) and inflammatory markers may be expected.

Renal impairment, with an eGFR less than 60mL/min may predispose a patient to gout. Raised Ca may be an indication of pseudo-gout, especially if the gout is non-responsive to allopurinal (which blocks urate production). A comparison of the crystal structure will provide a helpful differential.

16

Liver function tests

The liver has three key functions: storage (◀▶ deficiencies in iron, B_{12} and glucose); metabolic production (◀▶ production of cholesterol, CRP, fibrinogen (PT, INR, aPTT) and albumin); and detoxification of endogenous and exogenous toxins. Liver function tests (LFTs) will usually report enzymes, albumin and bilirubin levels. If the test name ends in 'ase' or has the units (IU/L) then it is an enzyme. An enzyme takes a substrate (usually not soluble and toxic) and converts it into a product (usually more soluble and less toxic).

Almost all enzymes are intracellular. The presence of high concentrations of enzymes in the blood means that cells have been destroyed, allowing the enzymes to 'leak out'. Possible causes of cellular destruction could be trauma, alcoholic cirrhosis, viral damage from hepatitis, or cellular necrosis (cell death) resulting from drug toxicity. The particular enzymes raised will usually indicate which type of damage has occurred.

A standard LFT may contain:

- Bilirubin: Plumbing of the liver – pre-, actual, post- and red blood cell turnover
- Alanine aminotransferase (ALT): Viral hepatitis and drug toxicity – refer to British National Formulary (see Further Reading, page 103) and the pharmacy department in your local healthcare setting
- Aspartate aminotransferase (AST): Alcoholic hepatitis, acute liver failure
- Alkaline phosphatase (ALP or Alk Phos): The biliary tree, gall stones, pancreas
- Gamma-glutamyltransferase (GGT): Alcohol, analgesics and opiates
- Amylase: Pancreatitis
- Albumin: Decreased on liver failure
- International normalised ratio (INR): Now being used as a surrogate liver marker. In liver failure, fibrinogen production is compromised, and clotting time (INR) is therefore significantly extended.

As the liver is a dynamic and interconnected organ, the blood results from the LFT may be difficult to interpret when presented as a list on a results screen or printout. Instead, it may

be helpful to imagine them superimposed onto a general anatomy and physiology diagram, and interconnected to other organs.

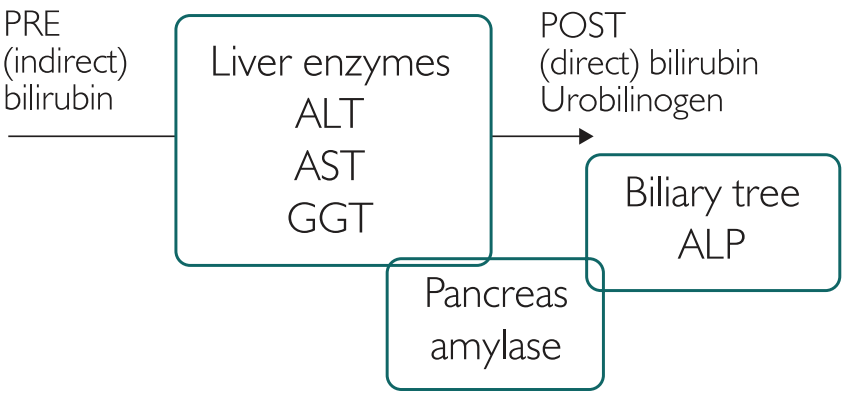


Figure 16.1: Liver enzymes

Bilirubin

Bilirubin is a breakdown product of red blood cells. Elevated bilirubin is called jaundice, and bilirubin is a marker of 'plumbing' in the liver. Pre-hepatic bilirubin is called indirect or unconjugated. In the liver an enzyme called UGT1A1 (which is dysfunctional in Gilbert's syndrome) converts indirect bilirubin into direct, or conjugated bilirubin. The conjugated bilirubin is then partly used to produce urobilinogen and partly excreted in the faeces. The LFT result of 'Total bilirubin' is direct + indirect, both of which can also be measured to further investigate liver dysfunction. As indirect bilirubin rises, this is usually a pre- or actual hepatic condition; a rise in the direct form usually indicates a post-hepatic blockage.

Jaundice can occur when high levels of bilirubin are produced in polycythaemia (◀▶ full blood count and red blood cell count). High levels of bilirubin from the red cells overload the liver and may predispose the patient to jaundice. Jaundice can also be seen in actual liver damage, with high liver enzyme concentrations of aspartate aminotransferase (AST), alanine aminotransferase (ALT) and gamma-glutamyltransferase (GGT). Often bilirubin rises before the liver enzymes do. This may occur in patients undergoing chemotherapy, or those with hepatitis (both viral and alcoholic), or being treated with antibiotics or with some genetic conditions. In a post-hepatic blockage (with high Alk phos/amylase and raised liver enzymes) bilirubin will increase and this can be linked to the absence of urobilinogen.

Aminotransferases

Alanine aminotransferase (ALT) is found inside liver cells (see the earlier chapter on ways in which blood can change). Therefore, as the liver cells are damaged, the ALT will leak out and we can now measure it in the blood. ALT is a liver enzyme that closely reflects chronic and acute liver damage. It is often raised in viral hepatitis, drug toxicity or overdose.

Aspartate aminotransferase (AST) is a liver enzyme that indicates acute liver damage. It is often raised in alcoholic patients, due to severe liver damage. It can be used in conjunction with ALT to assess the extent of liver dysfunction.

In a raised ALT, you should ask the following:

- Is the value more than 1.5 x above the upper range?
- Is the patient symptomatic?
- Are bilirubin, INR and albumin also abnormal?

If the answer to any of these questions is 'yes', further investigations should be considered. If the answer is 'no', then consider repeating the test.

In the following examples, the upper reference range for AST is 40 IU/L and for ALT 40 IU/L. The figures used demonstrate which enzyme is raised, and should not be used as a diagnostic cut-off in practice. Some surgeries use a clinical decision limit protocol of 1.5x to 2x as the upper limit for defining group A. Assuming the patient is asymptomatic, hasn't started a new medication, and the other liver markers are within a normal range, an ALT of less than 80 IU/L could perhaps be filed as a false positive.

Patient 1 has AST 100 IU/L and ALT 22 IU/L, and all other LFTs are normal. AST is also found in cardiac tissue so investigation of chest pain may reveal a cardiac event.

Patient 2 has ALT 100 IU/L and AST 48 IU/L, which could indicate viral hepatitis, toxicity to methotrexate, or over-use of analgesics.

Patient 3 is an alcoholic patient with severe liver damage. AST is 200 IU/L and ALT is 100 IU/L. If the AST was to increase to 500 IU/L the prognosis would become increasingly poor.

Patient 4 has obstetric cholestasis, presenting with pruritus (itch) and raised ALT. Bile salts are ordered. Bile salts are made by the liver and stored in the gall bladder; they are then secreted into the duodenum to dissolve fats (enterohepatic circulation). In hepatobiliary conditions, the enterohepatic circulation is dysfunctional and bile salts can be secreted into the systemic circulation. Given the lipid-dissolving, surfactant properties of bile salts, their presence in a pregnant patient's systemic circulation could lead to foetal distress or damage.

Gamma-glutamyltransferase (GGT)

The enzyme gamma-glutamyltransferase (GGT) is often used as a marker for alcohol intake. GGT can be elevated for up to five days following alcohol intake. However, ◀▶ full blood count (FBC) and the mean cell volume (MCV) because a raised GGT could be due to transient alcohol intake. A high GGT and a folate-deficient macrocytic anaemia, caused by alcohol intake, are more indicative of alcohol abuse. As this will take at least 6–12 weeks to develop, consider the red blood cell lifespan.

Alkaline phosphatase

The enzyme alkaline phosphatase (Alk phos or ALP) is also present in other organs (bones, kidney and placenta) so ◀▶ these tests, symptoms and history. Liver-specific Alk phos is an enzyme mainly found in the cells that line the bile ducts and biliary tree. Therefore ALP is usually elevated with bile duct blockage – caused by gall stones, pancreatitis, post-hepatic tumour or cholestasis.

If ALP is raised and GGT is not, then consider a non-hepatic condition. If GGT is raised, and/or ALP is twice the upper range, and/or both ALP and GGT have been raised for more than three months, and/or the patient has symptoms, consider a liver ultrasound.

Amylase

The enzyme amylase is found in the pancreas and saliva. It is therefore often elevated in pancreatitis.

Patient 5 had a rise in amylase (pancreas), bilirubin (post-hepatic blockage) and Alk phos (biliary tree), followed by GGT, ALT and then AST (secondary liver damage), which represented the progression of a pancreatic tumour.

Albumin

Albumin is a protein made by the liver and has three main roles: as a chaperone (transporter) for molecules like calcium (◀▶ bone profile); to provide pressure and osmotic stability in the blood, and is thus linked to oedema and renal function (◀▶ U&Es); and as a precursor for antibody production (globulins). Albumin can also be decreased in malnutrition, but other LFTs are likely to be normal. Since albumin is made by the liver, any dysfunction will probably reduce albumin concentrations (hypoalbuminaemia). However, you should check the U&Es as some renal dysfunction will not retain albumin, and proteins and cells are likely to be present in the urine.

Alcohol and drug abuse, and viral infection

Patient 6 has raised aminotransferases (>300 IU/L), due to a viral infection and paracetamol overdose, in addition to an alcoholic liver disease.

As the viral infection becomes acute, this value may rise higher still. An AST >400 IU/L and a peak ALT of 1000 IU/L is associated with severe liver damage due to paracetamol overdose. For the hepatitis viral infection in practice, this can also be seen in the diagnosis of viral hepatitis. Hepatitis B surface antigen (HepBsAg) may indicate a current infection. Alternatively, a new, rapid technique is to measure the DNA or RNA of the HepB virus directly in the cell.

Patients who use cocaine, marijuana or heroin alone will usually have out-of-range LFTs, including AST and GGT. If injecting, a raised ALT may often indicate a viral hepatitis. If the patient is alcoholic (raised MCV and raised GGT), additional drug use will also affect these measures.

Storytelling: ALT: Imagine a car engine (the liver). The engine contains oil (ALT). If you notice some oil on the driveway under the car (ALT in the blood), then it's likely that the oil has leaked out of the engine. The more oil has leaked out, the more damaged the engine is. Likewise, the more ALT there is in the blood, the more ALT must have leaked out of the liver cells. This is an example of cellular content. ALP: This enzyme is also found in the cells after they have been in the liver – any damage in the liver will make the ALP leak out into the blood. The ALP level can therefore give us information about a post-hepatic or obstructive jaundice. This is another example of cellular content. Bilirubin: When a red blood cell reaches the end of its lifespan, it is recycled and a single bilirubin molecule is released into the blood. These single bilirubin molecules are known as 'unconjugated' bilirubin. The liver has a 24 hour 'dating agency' which pairs up single bilirubin molecules. These paired bilirubin molecules are 'conjugated' and they are more easily excreted. We can find out if the patient has a pre- or post-hepatic jaundice by looking at the type of bilirubin. If this information isn't available, we can look at associated tests. A pre-hepatic jaundice will usually also have raised red blood cells. An actual liver damage jaundice may have raised ALT and a post-hepatic jaundice may also have raised ALP.

Afterword

Thomas Edison once said, 'The object of all work is production or accomplishment and to either of these ends there must be forethought, system, planning, intelligence, and honest purpose, as well as perspiration'; or as Bane (a supervillain from Batman) also said, 'all that matters is having a plan'.

I hope that this book has helped to formulate a plan, to create a system, to encourage the creation of a protocol, a pattern. We have explored family groups, some questions to ask when results are out of range, and how different blood components change with pathology. We have introduced storytelling as a way of explaining and remembering complex and abstract concepts. Good luck. Have fun with blood tests (yes, they can be fun) and let me know how it's going at **@grahambasten** on Twitter.

Good luck!

Further reading and references

There are lots of great resources on the internet. Try searching for the following, as good examples:

Arthritis Research UK

BD Vacutainer Tube Guide

British Committee for Standards in Haematology

British National Formulary

British Thyroid Foundation

Camden CCG Blood Protocol

CSP First Contact Diabetes UK

European Commission Public Health

Health and Care Professions Council

Health and Social Care Board Northern Ireland

Institute of Biomedical Science

Lab Tests Online

Leeds PathologyLeicester Diabetes Pathway

Manchester Anaemia Pathway Midline Plus Blood Tests (USA)

NHS – Blood tests

NHS Choices

NHS Guidelines NHS Inform Scotland

NHS PathwaysNursing and Midwifery Council

Nursing Times

Public Health England Fingertips

Public Health England SHAPE Royal College of Nursing

Scottish Health Council Digital Stories

Index

abnormal results, interpreting *35–40*
acid base *19, 75–78, 89*
acid base derangements *77*
acid base measures *76*
acidosis, metabolic *77*
acidosis, respiratory *76, 77*
Acute Kidney Injury (AKI) blood report *90*
Addison's disease *88, 90*
adhesion molecules *53*
alanine aminotransferase (ALT) *17, 21, 95, 97*
albumin *18, 86, 95, 98*
alcohol *51, 97, 99*
alcoholic cirrhosis *95*
alcoholic liver disease *99*
alcoholic neuropathy *45*
alkaline phosphatase *17, 23, 85, 95, 98*
alkalosis, metabolic *77*
alkalosis, respiratory *77*
allopurinal *93*
alpha-1-antitrypsin *76*
aminotransferases *97, 99*
amylase *18, 95, 98*
anaemia *30, 38, 43, 44, 45*
anaemia, aplastic *47*
anaemia of chronic disease *45*
anaemia, pernicious *44, 60*
angina *73*
ankylosing spondylitis (AS) *60, 61*
antibiotics *96*
anticoagulants *6, 7*
anti-inflammatory treatments *58*
anti-neutrophil cytoplasmic antibodies (ANCA) 61
aspartate aminotransferase (AST) *17, 21,73, 95, 97*
aspirin *51*
autoimmune conditions *41, 47, 59–63*
autoimmune markers *19*

basophil *15, 47*
Bence Jones protein *18, 47, 62*
bile salts *97*
bilirubin *18, 23, 95, 96*
bisphosphonates *85*
blast cells *15, 47*
blood collection and storage techniques *5, 86*
blood group *65*
blood tests, terms used in *14*
bone marrow failure *41*
bone profile *18, 83–86*
bone turnover *83, 84*
B-type natriuretic peptide (BNP) *20, 73*

CA-125 *19*
calcitriol *83*
calcium *86, 93*
carbimazole *81*
cardiovascular disease (CVD) *71*
cardiovascular risk *72*
cardiovascular risk in diabetes *69*
cell damage, response to *26*
cell leakage *21*
cellular content *21, 99*
cellular inflammation *53, 54*
cellular signalling molecules *53*
chemotherapy *47, 96*
cholesterol *71–73*
cholesterol treatments *72*
chronic fatigue syndrome *63*
chronic obstructive pulmonary disease (COPD) *42, 43, 73, 75–78, 88*
chylomicrons *71*
clexane *50*
clinical cut-off values *10*
clinical decision limits (CDL) *10*
clinical sensitivity and specificity *9*
clopidogrel *51*
coagulation *49–52*
coagulation monitoring *50*
congestive heart failure *88*
Conn's syndrome *88*
coronary heart disease (CHD) *71*
corrected Ca *86*
C-reactive protein (CRP) *13, 16, 54, 57*
creatine kinase mb (CKmb) *20, 22, 73*
creatinine *17, 23, 90*
cyclo-oxygenase 2 (COX-2) *53, 54*
Cushing's disease *88*

D-Dimer *18, 49*
deep vein thrombosis (DVT) *18, 49–52*
deltaparin *50*
dementia *46*
demyelination *22*
diabetes *67–70*
diabetes diagnosis and treatment *69*
diabetes insipidus *67*
diabetes mellitus *67*
diabetic complications *68*
diabetic ketoacidosis *69*
diabetic neuropathy *45, 69*
diabetic renal impairment *69*
diabetic retinopathy *69*
drug abuse *99*
drug toxicity *97*

elderly patients *88*
endometriosis *91*
enzyme *95*
eosinophil *15, 47*
erythrocyte sedimentation rate (ESR) *16, 53, 55, 56*
erythropoiesis *42*
erythropoietin *24, 25, 42, 75, 87*
estimated glomerular filtration rate (eGFR) *17, 23, 43, 75, 83, 91, 92*
exenatide *70*
exudative inflammation *53, 54*

ferritin *19, 44*
fibrinogen *50*
fibromyalgia *63*
foam cells *47*
folate *19, 43, 44*
full blood count *29, 41–48*

gamma-glutamyl transferase (GGT) *17, 95, 98*
Gilbert's syndrome *96*
globulin *18, 37*
glomerular filtration rate *23, 96*
glomerular nephritis *61*
glucose *68*
glucose measurement *69, 70*
glycogen *68*
gout *45, 92*
granulomatosis *61*

H ions, concentration of *76*
haematocrit (Hct) *15, 30, 37, 46*
haemoglobin (Hb) *15, 30, 41*
haemoglobin with glucose irreversibly bound (HbA1c) *15, 19, 70*
haemolysed samples *7*
haemophilic conditions *52*
heparin *50*
hepatitis screening *66*
hepatitis *97*
hepatitis, viral *60, 95, 97, 99*
high-density lipoprotein (HDL) *71*
Hodgkin's lymphoma *47*
homocysteine *45*
human immunodeficiency virus (HIV) *47, 60*
hypercalcaemia *86*
hyperkalaemia *77, 89*

hypernatraemia *87*
hyperparathyroidism *86*
hyperthyroidism *80*
hypocalcaemia *85, 86*
hypokalaemia *89*
hyponatraemia *88*
hypothyroidism *80*
hypoxia 42

ileostomy *89*
immune system *59–63*
infection *41, 47, 99*
inflammatory markers *53–58*
input/output linked systems *33*
insulin *68, 89*
insulin-mediated storage of glucose as glycogen 68
interaction *27*
international normalised ratio (INR) *18, 50, 95*
iron *19, 43*
iron deficiency *43*

jaundice *23, 24, 96, 99*

kidney *22, 42, 83, 89*
kidney disease, chronic *85, 92*
kidney function *17, 87–93*

leukaemia *41, 47*
leukopenia *47*
liquorice *89*
liver damage *86, 97, 99*
liver enzymes *12, 21, 96*
liver failure *88*
liver function *23, 24, 50, 95*
liver function tests (LFTs) *17, 31, 32, 95–99*
low-density lipoprotein (LDL) *71*
low molecular weight heparin *50*
lymphocyte *15, 31, 47*
lymphoma *47*

magnesium *85*
malnutrition *86*
mean cell volume (MCV) *15, 43, 44*
metformin *70*
methotrexate *38*
monocyte *15, 47*
myeloma *47, 62*
myocardial infarction (MI) *73*
myositis *73*
nephrotic syndrome *88*
network diagram *34*
neuropathy *45*
neutrophils *15, 30, 37, 47, 55*

oestrogen *83*
order of draw *7*
osteoblasts *83*
osteoclasts *83*
osteocyte *83*
osteomalacia *85*
osteoporosis *83, 84*
overdose *97*

Paget's disease *96*
paracetamol overdose *99*
parathyroid hormone (PTH) *83, 86*
phlebotomy technique *5, 89*
phosphate *86*
plasma *5*
plasma and serum, difference between *5*
plasma viscosity (PV) *16, 37, 53, 55, 57*
platelet *14*
pneumonia *88*
polycythaemia *42, 46, 75, 96*
polymyalgia rheumatica (PMR) *62*
polymyositis *62*
polyuria *67, 68*
potassium (K) *17, 22, 89*
prostate specific antigen (PSA) *19*
psoriatic arthritis (PA) *62*
pulmonary embolism (PE) *18*
purines *93*

reactive arthritis *62*
red blood cell count (RBC) *15, 30, 75*
red blood cell indices *41*
red blood cell production *25*
reference ranges *4, 5, 8, 11*
renal anaemia *25, 43*
renal function *87–93*
renal salt-wasting *88*
renin-angiotensin system *87*
reticulocytes *42*
rhabdomyolysis *73*
Rhesus (Rh) status *65*
rheumatoid arthritis (RA) *35, 37, 38, 41, 60, 61*
rickets *85*

seropositive/seronegative *60, 61*
serum *5*
sodium (Na) *17, 87*
statins *62, 72*
syndrome of inappropriate anti-diuretic hormone (SIADH) *88*
systemic lupus erythematosus (SLE) *60, 61*

T3 *79,*
thrombocythaemia *50*
thrombocytopenia *14, 61*
thyroid function *19, 79–81*
thyroid hormones *80*
thyroid-stimulating hormone (TSH) *79, 80*
thyroid treatments *81*
thyrotropin-releasing hormone (TRH) *79, 80*
thyroxine (T4) *79, 80*
total iron binding capacity (TIBC) *45*
transfusion testing *65–66*
triglycerides *72*
troponins *20, 22, 73*
tuberculosis *76*

units *4*
urate *92*
urea *22, 90*
urea:creatinine ratio *91*
urea and electrolytes (U&Es) *17, 67, 87–93*
uric acid *92*
urobilinogen *18*

values outside range, strategy for *11*
vasculitis *61*
vasopressin *67, 88*
venous thromboembolism (VTE) *49*
viral infection *99*
viral infection, normal response to *59*
vitamin B12 *19, 43, 44*
vitamin D3 *83, 87*
von Willebrand's factor (VWF) *51*

warfarin *50*
waste products, clearance of *22*
Well's score (WS) *49*
white blood cell count (WBC) *14, 26, 30, 37, 47, 56, 75, 93*
white blood cell indices *46*
white blood cell production *25*